

EFFECTS OF DIETARY ENZYMES OR
SPECIALTY PROTEINS ON NURSERY PIG PERFORMANCE

by

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Abstract

Eight experiments used 1,712 pigs to determine influences of dietary enzymes or specialty proteins on nursery pig performance. Experiments 1 and 2 evaluated the effects of fish meal, fermented soybean meal, or dried porcine solubles on performance. Experiment 1 showed pigs fed dried porcine solubles had improved ($P = 0.01$) ADG and G:F compared to pigs fed the control diet, and improved ($P = 0.03$) G:F compared to pigs fed the combination of fermented soybean meal and fish meal. Experiment 2 showed pigs fed increasing fermented soybean meal had improved (quadratic, $P = 0.03$) G:F. Experiments 3 and 4 evaluated the effects of commercial enzyme addition to diets containing dried distillers grains with solubles (DDGS) on performance. In experiment 3, neither DDGS nor enzyme addition influenced ($P > 0.10$) ADG and G:F. Experiment 4 found there were no ($P > 0.32$) enzyme \times DDGS source interactions. Corn DDGS did not influence pig performance ($P > 0.36$). Sorghum DDGS reduced ($P = 0.003$) G:F, with no difference between sorghum DDGS sources. Adding enzymes to 30% DDGS diets did not improve ($P > 0.57$) performance. Experiments 5 and 6 evaluated the effects of fish meal (SMFM), spray-dried animal plasma (SDAP), or peptone on performance. In Experiment 5, different specialty proteins had similar ($P > 0.10$) ADG, ADFI, or G:F. Experiment 6 showed pigs fed 4% Peptone 2 during phase 1 and 2% Peptone 2 during phase 2 had improved ($P < 0.05$) ADG compared to pigs fed SMFM, and improved ($P < 0.05$) G:F compared to pigs fed all other diets. Experiments 7 and 8 developed an available P release curve for commercial phytase products. In both experiments, pigs fed increasing inorganic P had improved (linear, $P < 0.01$) G:F and percentage bone ash. Pigs fed increasing OptiPhos 2000-M, Phyzyme XP, or Ronozyme P had improved ($P < 0.001$) percentage bone ash. Available P release for up to 1,000 FTU/kg of *Escherichia coli*-derived phytases can be predicted by the equation ($y = -0.000000125x^2 + 0.000236245x + 0.015482$), where x is the phytase level in the diet.

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CHAPTER 1 - Effects of fermented soybean meal and specialty animal protein sources on nursery pig performance

Abstract

In 2 experiments, 602 pigs were used to evaluate the effects of dietary fish meal, fermented soybean meal, or dried porcine solubles on nursery pig performance. In Exp. 1, 252 nursery pigs (PIC TR4 × 1050, initial BW 6.8 kg) were fed either (1) a control diet containing no specialty protein sources or the control diet with (2) 5% fish meal, (3) 3.5% dried porcine solubles, (4) 6.0% fermented soybean meal, (5) a combination of 1.75% fermented soybean meal and 1.75% dried porcine solubles, or (6) 3.0% fermented soybean meal and 2.5% fish meal. There were 7 replications with 6 pigs per pen. Experimental diets were fed from d 0 to 14; then all pigs were fed a common diet for 14 d without specialty protein sources. From d 0 to 14, pigs fed dried porcine solubles alone or with fermented soybean meal had improved ($P < 0.05$) ADG and G:F compared with pigs fed all other diets. Overall (d 0 to 28), pigs fed dried porcine solubles had improved ($P = 0.01$) ADG (421 vs. 383 g) and G:F (0.77 vs. 0.73) compared with pigs fed the control diet and improved ($P = 0.03$) G:F (0.77 vs. 0.74) compared with pigs fed the combination of fermented soybean meal and fish meal. In Exp. 2, 350 weanling pigs (PIC C22 × 1050, initial BW 6.1 kg) were fed either (1) a control diet containing no specialty protein sources or the control diet with (2) 3% fish meal, (3) 6% fish meal, (4) 3.75% fermented soybean meal, (5) 7.50% fermented soybean meal, (6) a combination of 1.88% fermented soybean meal and 1.88% dried porcine solubles, or (7) a combination of 3.75% fermented soybean meal and 3.75% dried porcine solubles. There were 10 replications with 5 pigs per pen. Experimental diets were fed from d 0 to 14, and then all pigs were fed a common diet for 21 d without specialty protein sources. From d 0 to 14, pigs fed increasing fish meal had increased (quadratic $P = 0.05$) ADFI. Pigs fed increasing fermented soybean meal had improved (quadratic, $P = 0.01$) G:F. Pigs fed the combination of fermented soybean meal and dried porcine solubles had improved ($P = 0.05$, $P = 0.03$) ADG and G:F, respectively, compared with pigs fed diets containing fish meal and improved ($P = 0.01$, $P = 0.02$) ADG and ADFI, respectively, compared with pigs fed diets containing fermented soybean meal. Overall (d 0 to 35), pigs fed increasing fermented soybean

meal had improved (quadratic, $P = 0.03$) G:F. In conclusion, these 2 trials show that feeding nursery pigs dried porcine solubles alone or with fermented soybean meal can improve growth performance.

Key words: dried porcine solubles, fermented soybean meal, growth, nursery pig

Introduction

Several studies show that fish meal improves growth and immune system function in nursery pigs (Kim and Easter, 2001; Young et al., 2002; Gaines et al., 2005). However, responses to fish meal tend to be inconsistent because of its variability (Wiseman et al., 1991). Plant proteins, such as soybean meal, are less expensive than animal protein sources, but contain anti-nutritional factors that are not suitable to be fed as the sole protein source post weaning (Li et al., 1990, 1991; Friesen et al., 1993; Qin et al., 1996). Research has indicated that pigs fed fermented, rather than solvent-extracted, soybean meal have improved feed efficiency and amino acid digestibility (Min et al., 2004; Kim et al., 2007; Cho et al., 2008). The fermentation process is thought to eliminate residual trypsin inhibitors and some oligosaccharides in soybean meal that may decrease pig performance.

Another possible protein source for nursery diets is dried porcine solubles, a coproduct of the heparin (a human pharmaceutical product) industry. The solubles are made from porcine intestinal mucosa and contain a high level of digestible peptides and amino acids (Maxwell and Carter, 2001). Newly weaned pigs have a considerable capacity to absorb peptides in the small intestine (Gilbert et al., 2008). Dried porcine solubles have previously been shown to improve growth performance of nursery pigs, possibly because the product supplies a high level of these small peptides (Zimmerman et al., 1997; Lindemann et al., 1998; Carter et al., 1999; DeRouchey et al., 2003).

Although the positive effects of dried porcine solubles have been demonstrated in nursery pigs, less information is available on fermented soy products or the combined use of these protein products. Therefore, the objective of these experiments was to evaluate the effects of fish meal, fermented soybean meal, and dried porcine solubles on growth performance of weanling pigs.

Materials and methods

General

Experimental procedures were approved by the Kansas State University Animal Care and Use Committee (number 2461). Specialty proteins used in Exp. 1 and Exp. 2 were from the same batches. They were sampled and analyzed for CP and AA (AOAC, 2000; Table 1).

Experiment 1

A total of 252 pigs (TR4 \times 1050, PIC Hendersonville, TN; initial BW 6.8 kg) were used in a 28-d growth trial to evaluate the effects of fish meal, fermented soybean meal, and dried porcine solubles on nursery pig performance. Pigs were blocked by weight and allotted to 1 of 6 dietary treatments. There were 6 pigs per pen and 7 pens per treatment. Each pen (1.2 m²) contained 1 self-feeder and 1 nipple waterer to provide ad libitum access to feed and water. Pigs were housed in the Kansas State University Swine Teaching and Research Center in Manhattan.

A common pelleted starter diet was fed from weaning until the start of the experiment (d 7). The 6 experimental treatments were: (1) negative control, (2) 5% fish meal, (3) 3.5% dried porcine solubles (DPS 50, Nutra-Flo, Sioux City, IA), (4) 6.0% fermented soybean meal (PepSoyGen, Nutra-Flo, Sioux City, IA), (5) 1.75% fermented soybean meal and 1.75% dried porcine solubles, and (6) 3.0% fermented soybean meal and 2.5% fish meal (Table 2). Treatments 2 through 6 were formulated with the same dietary soybean meal level (31.4%). Because standard standardized ileal digestible values were not available on the fermented soybean meal product evaluated, diets were formulated on a total AA basis. Treatment diets were fed for 14 d; then, all pigs received a common diet for 14 d. All diets were in meal form. Average daily gain, ADFI, and G:F were determined by weighing pigs and measuring feed disappearance on d 7, 14, 21, and 28 of the trial.

Experiment 2

A total of 350 pigs (C22 \times 1050, PIC Hendersonville, TN; initial BW 6.1 kg) were used in a 35-d growth trial to evaluate the effects of fish meal, fermented soybean meal, and the combination of fermented soybean meal and dried porcine solubles on weanling pig performance. Pigs were blocked by weight and allotted to 1 of 7 dietary treatments. There were 5 pigs per pen and 10 pens per treatment. Each pen (1.5 m²) contained 1 self-feeder and 1 cup

waterer to provide ad libitum access to feed and water. The study was conducted at the Kansas State University Segregated Early Weaning Facility in Manhattan.

A common pelleted starter diet was fed from weaning until the start of the experiment (d 7). The 7 dietary treatments were: (1) negative control diet or the control diet with (2) 3% fish meal, (3) 6% fish meal, (4) 3.75% fermented soybean meal, (5) 7.50% fermented soybean meal, (6) 1.88% fermented soybean meal and 1.88% dried porcine solubles, and (7) 3.75% fermented soybean meal and 3.75% dried porcine solubles (Table 3). Treatment diets 2, 4, and 6 were each formulated with 35.7% soybean meal; diets 3, 5, and 7 each had 29.8% soybean meal. Because standard standardized ileal digestible values were not available on the fermented soybean meal product evaluated, diets were formulated on a total AA basis. Treatment diets were fed for 14 d; then, all pigs received a common diet for 21 d. All diets were in meal form. Average daily gain, ADFI, and G:F were determined by weighing pigs and measuring feed disappearance on d 7, 14, 24, and 35 of the trial.

Statistical Analysis

Data were analyzed as a randomized complete block design with pen as the experimental unit. Data were analyzed by using an ANOVA in the MIXED procedure of SAS with the weight block as a random effect and treatments as a fixed effect. All possible pairwise comparisons were used to evaluate differences among treatments in Exp 1. In Exp 2, preplanned linear and quadratic contrasts were used to determine the effects of increasing levels of fish meal, fermented soybean meal or combination of fermented soybean meal and dried porcine solubles. For all contrasts the negative control diet without the protein source evaluated was used as the first dose level. Additional contrasts were used to compare the mean of the effects of fish meal or fermented soybean meal addition versus the effects of inclusion of the combination of fermented soybean meal and dried porcine solubles. Means were considered significant if their *P*-values were < 0.05 and trends if their *P*-values were < 0.10.

Results

Crude protein and AA analysis of the specialty protein sources were generally consistent with values supplied by the manufacturer that were used in diet formulation (Table 1). However, the analyzed Lys level for dried porcine solubles was significantly higher than formulated levels (3.81% vs. 3.10%).

Experiment 1

From d 0 to 14, pigs fed dried porcine solubles alone or in combination with fermented soybean meal had improved ($P < 0.05$) ADG compared with pigs fed all other diets (Table 4). Pigs fed dried porcine solubles tended to have increased ($P < 0.10$) ADFI compared with pigs fed the negative control diet or the diet containing the combination of fermented soybean meal and fish meal. Pigs fed dried porcine solubles with or without fermented soybean meal had improved ($P < 0.05$) G:F compared with pigs fed the negative control diet or diets containing fish meal with or without fermented soybean meal. Pigs fed dried porcine solubles had improved ($P < 0.05$) G:F compared with pigs fed fermented soybean meal. Finally, pigs fed the combination of fermented soybean meal and dried porcine solubles tended to have improved ($P < 0.10$) G:F compared with pigs fed fermented soybean meal alone.

During the 14-d period when pigs were fed a common diet, pigs previously fed fermented soybean meal alone or in combination with fish meal had increased ADG ($P < 0.05$), and pigs previously fed fish meal alone tended to have increased ($P < 0.10$) ADG compared with pigs fed the combination of fermented soybean meal and dried porcine solubles. Pigs previously fed the combination of fermented soybean meal and fish meal also tended to have increased ($P < 0.10$) ADFI compared with pigs fed the combination of fermented soybean meal and dried porcine solubles.

Overall (d 0 to 28), pigs fed dried porcine solubles had improved ($P < 0.05$) ADG compared with pigs fed the control diet and tended to have improved ($P < 0.10$) ADG compared with pigs fed the diet containing the combination of fermented soybean meal and fish meal. Pigs fed fermented soybean meal alone also tended to have increased ($P < 0.10$) ADG compared with pigs fed the control diet or the diet containing the combination of fermented soybean meal and fish meal. Pigs fed dried porcine solubles alone had improved ($P < 0.05$) G:F compared with pigs fed the control diet or the combination of fermented soybean meal and fish meal and tended to have improved G:F compared with pigs fed only fish meal. Pigs fed diets containing the combination of fermented soybean meal and dried porcine solubles also tended to have improved ($P < 0.10$) G:F compared with pigs fed the control diet.

Experiment 2

From d 0 to 14, pigs fed increasing levels of fish meal tended to have increased (quadratic, $P = 0.08$; linear, $P = 0.07$) ADG and G:F, respectively, and had increased (quadratic $P = 0.05$) ADFI (Table 5). Feeding increasing levels of fermented soybean meal did not influence ($P > 0.10$) ADG or ADFI but did improve (quadratic, $P = 0.01$) G:F. Pigs fed increasing levels of the combination of fermented soybean meal and dried porcine solubles tended to have improved (linear, $P = 0.06$) ADG and had improved ($P = 0.002$) G:F. In addition, pigs fed the combination of fermented soybean meal and dried porcine solubles had improved ($P = 0.05$, $P = 0.03$) ADG and G:F, respectively, compared with pigs fed diets containing fish meal and improved ($P = 0.01$, $P = 0.02$) ADG and ADFI, respectively, compared with pigs fed diets containing fermented soybean meal.

There were no significant main effects or treatment differences for ADG or ADFI ($P > 0.16$) from d 14 to 35 (common period). Overall (d 0 to 35), pigs fed increasing levels of fermented soybean meal had improved (quadratic, $P = 0.03$) G:F, and pigs fed increasing levels of the combination of fermented soybean meal and dried porcine solubles tended to have improved (linear, $P = 0.06$) G:F.

Discussion

Plant protein products, such as soybean meal, are generally less expensive than animal protein sources, but may contain anti-nutritional factors for newly weaned nursery pigs (Li et al., 1990, 1991; Friesen et al., 1993; Qin et al., 1996). Newly weaned pigs often experience hypersensitivity to soy protein but begin to develop tolerance after 7 to 10 days (Barratt et al., 1978). Other proteins, especially from animal-based sources that are highly digestible are commonly fed to stimulate ADFI and ADG in nursery pigs postweaning to help avoid this period of transient hypersensitivity.

Fish meal is a high-protein feed ingredient with a desirable amino acid profile for nursery pig diets. Church and Kellems (1998) showed that fish meal contains the amino acids that are generally deficient in cereal grains in addition to vitamins and minerals that are often deficient in other protein sources. In particular, fish meal contains high levels of sulfur amino acids. Kim and Easter (2001), Young et al. (2001), and Gaines et al. (2005) showed that dietary fish meal increased both growth performance and resistance to disease in nursery pigs. However, Wiseman

et al. (1991) showed that fish meal is highly variable because of the quality of fish and processing factors. The published analysis of Special Select menhaden fish meal (Kim and Easter, 2001; Young et al., 2001) showed differences of 0.26% in Lys (4.74 vs. 4.48), 0.15% in Met (1.77 vs. 1.62), 0.12% in Thr (2.55 vs. 2.43), and total AA (59.03% vs. 57.26%). These experiments showed inconsistency in growth responses, even when feeding the same batches of fish meal. An inconsistent response was also found in our studies, in which nursery pigs fed 3% fish meal tended to have increased ADG and had increased ADFI compared with pigs fed 6% fish meal from d 0 to 14. However, neither fish meal treatment resulted in pigs with improved growth performance compared with pigs fed the control diet. Bergstrom et al. (1997) showed that pig health status can affect the level of growth response in pigs fed fish meal. Disease-challenged pigs have a greater response to fish meal than healthier pigs. Pigs in our studies were of high health, which may help explain some of the variation in response to diets containing fish meal.

Another animal-based protein product that has shown a more consistent improvement in growth in nursery pigs is dried porcine solubles (Zimmerman et al., 1997; Lindemann et al., 1998; Carter et al. 1999; DeRouchey et al., 2003). Our data confirmed earlier research in that pigs fed dried porcine solubles had improved ADG compared with pigs fed all other diets and improved ADFI and G:F compared with pigs fed the control diet.

To further develop diets without animal-based protein products and to achieve similar performance, new soybean meal processing technologies are continually being examined. It is believed that additional processing of the soy protein may decrease the hypersensitivity problems found when these proteins are fed to newly weaned pigs. Previous research suggests that fermented soybean meal products increase nutrient digestibility and growth performance in nursery pigs (Min et al., 2004; Kim et al., 2007; Cho et al., 2008). However, little work has been completed in North American-type diets. In our studies, pigs fed increasing fermented soybean meal alone had improved G:F in one study, whereas in the second study, pigs fed fermented soybean meal tended to have increased ADG compared with pigs fed the control diet. This is in agreement with research presented by Kim et al. (2007) and Cho et al. (2008), which showed that pigs fed increasing levels of fermented soybean meal had increased G:F. Cho et al. (2008) suggested that increased efficiency in pigs fed fermented soybean meal was due to improved nutrient digestibility, specifically His and Lys. Min et al. (2004) showed that pigs fed increasing levels of fermented soybean meal had increased ADG and improved His, Lys, and Thr

digestibility compared with pigs fed 5% spray-dried animal plasma. Similarly, Kim et al. (2007) found that pigs fed increasing levels of fermented soybean meal had improved ADG, ADFI, and G:F and concluded that this was due to pigs fed fermented soybean meal having increased digestibility in all nutrients except P. Cho et al. (2008) demonstrated that although feeding fermented soybean meal again increased feed efficiency and His and Lys digestibilities, ADG was not improved.

Although specialty protein sources can be fed individually in a complete diet, combining them may provide complimentary benefits. It has been suggested that feeding dried porcine solubles in addition to other protein sources may have additive effects on pig performance. For instance, Kim et al. (2001) found that including 3% dried porcine solubles with 6% spray dried porcine plasma protein maximized growth performance from d 0 to 21 post weaning. Our data show that feeding alternative protein sources in place of high levels of soybean meal or fish meal can be accomplished with dried porcine solubles, either alone or in combination with fermented soybean meal.

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Figures and Tables

Table 1. Analyzed nutrient composition (as-fed basis)

Item;	Fish meal		Fermented SBM ¹		Dried porcine solubles ²	
	Formulated ³	Analyzed ⁴	Formulated ⁵	Analyzed ⁴	Formulated ⁵	Analyzed ⁴
CP, %	62.90	65.34	54.25	56.37	50.00	51.01
AA, %						
Arg	---	3.74	---	3.92	---	2.72
His	---	1.35	---	1.45	---	1.06
Ile	2.57	2.53	1.80	2.69	1.80	2.06
Leu	4.54	4.46	3.40	4.55	3.40	3.94
Lys	4.81	4.74	3.20	3.46	3.10	3.81
Met	1.77	1.71	0.71	0.80	0.90	0.96
Phe	---	2.55	---	3.12	---	2.23
Thr	2.64	2.52	2.15	2.22	2.00	2.10
Trp	0.66	0.59	0.49	0.75	0.35	0.25
Val	3.03	2.96	2.32	2.83	2.40	2.60
Ala	---	4.06	---	2.57	---	2.95
Cys	0.57	0.47	0.97	0.78	0.85	0.78
Gly	---	4.82	---	2.47	---	3.65
Orn	---	0.08	---	0.08	---	0.32
Pro	---	2.94	---	2.98	---	2.83
Ser	---	2.19	---	2.55	---	1.86
Tau	---	0.47	---	0.03	---	0.20
Tyr	---	1.99	---	2.18	---	1.86

¹SBM = soybean meal; PepSoyGen (Nutra-Flo, Sioux City, IA).

²DPS 50 (Nutra-Flo, Sioux City, IA).

³Nutrient values from the 1998 NRC.

⁴Mean value of one sample analyzed in duplicate.

⁵Nutrient values provided by the manufacturer.

Table 2. Diet composition (Exp. 1, as-fed basis)

Item;	Control	Fish meal 5.00%	Dried porcine solubles ² 3.50%	Fermented SBM ³ 6.00%	Fermented SBM ² + dried porcine solubles ³ 1.75% + 1.75%	Fermented SBM ² + fish meal ³ 3.00% + 2.50%	Common
Ingredient, %							
Corn	45.45	49.97	50.30	47.93	50.25	48.95	61.27
Soybean meal, 46.5% CP	40.01	31.42	31.38	31.40	31.40	31.39	33.85
Select menhaden fish meal	---	5.00	---	---	---	2.50	---
Fermented soybean meal	---	---	---	6.00	1.75	3.00	---
Dried porcine solubles	---	---	3.50	---	1.75	---	---
Spray dried whey	10.00	10.00	10.00	10.00	10.00	10.00	---
Soybean oil	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Monocalcium phosphate, 21% P	1.53	0.90	1.38	1.55	1.45	1.23	1.65
Limestone	0.98	0.68	1.13	0.98	1.08	0.83	0.95
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.35
Vitamin premix ⁴	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace mineral premix ⁴	0.15	0.15	0.15	0.15	0.15	0.15	0.15
L-Lys·HCl	0.15	0.15	0.32	0.22	0.32	0.19	0.30
DL-Met	0.12	0.11	0.16	0.14	0.17	0.13	0.12
L-Thr	0.06	0.07	0.13	0.08	0.13	0.08	0.11
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Calculated composition ⁵							
Total Lys, %	1.53	1.53	1.53	1.53	1.53	1.53	1.42
Lys:ME ratio, g/Mcal	4.61	4.55	4.58	4.60	4.59	4.59	4.23
Total Met:Lys	31	33	33	32	33	33	31
Total Met & Cys:Lys	58	58	58	58	58	58	57
Total Thr:Lys	65	65	65	65	65	65	64
Total Trp:Lys	20	19	17	18	17	18	18
CP, %	23.9	23.5	22.3	23.5	22.4	23.5	21.4
ME, kcal/kg	3,325	3,358	3,340	3,325	3,333	3,340	3,347
Ca, %	0.88	0.88	0.88	0.88	0.88	0.88	0.80
P, %	0.80	0.77	0.77	0.80	0.77	0.78	0.75
Available P, %	0.47	0.47	0.47	0.47	0.47	0.47	0.42

¹Pigs were a common diet from weaning for 7 d, experimental diets were then fed for 14 d, followed by a 14 d common period.

²DPS 50 (Nutra-Flo, Sioux City, IA).

³SBM = soybean meal; PepSoyGen (Nutra-Flo, Sioux City, IA).

⁴Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B₁₂; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄·H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

⁵Values for fish meal were from NRC (1998) and values for fermented soybean meal and dried porcine solubles were provided by the manufacturer.

Table 3. Diet composition (Exp. 2; as-fed basis)¹

Item;	Control	Fish meal		Fermented SBM ²		Fermented SBM ² + dried porcine solubles ³		Common
		3.00%	6.00%	3.75%	7.50%	1.88% + 1.88%	3.75% + 3.75%	
Ingredient, %								
Corn	45.45	48.26	50.72	46.96	48.12	46.96	48.11	61.27
Soybean meal, 46.5% CP	40.01	34.76	29.87	34.65	29.66	34.70	29.76	33.85
Select menhaden fish meal	---	3.00	6.00	---	---	---	---	---
Fermented soybean meal	---	---	---	3.75	7.50	1.88	3.75	---
Dried porcine solubles	---	---	---	---	---	1.88	3.75	---
Spray dried whey	10.00	10.00	10.00	10.00	10.00	10.00	10.00	---
Soybean oil	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Monocalcium P, 21% P	1.53	1.15	0.78	1.55	1.55	1.45	1.35	1.65
Limestone	0.98	0.80	0.60	1.00	1.03	1.05	1.13	0.95
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.35
Vitamin premix ⁴	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace mineral premix ⁴	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
L-Lys·HCl	0.15	0.15	0.15	0.19	0.23	0.19	0.23	0.30
DL-Met	0.12	0.11	0.11	0.13	0.13	0.12	0.13	0.12
L-Thr	0.06	0.07	0.07	0.07	0.08	0.07	0.09	0.11
Total								
Calculated composition ⁵								
Total Lys, %	1.53	1.53	1.53	1.53	1.53	1.53	1.53	1.42
Lys:ME ratio, g/Mcal	4.61	4.57	4.55	4.61	4.60	4.58	4.58	4.23
Total Met:Lys	31	32	34	31	31	31	32	31
Total Met & Cys:Lys	58	58	58	58	58	58	58	57
Total Thr:Lys	65	65	65	65	65	65	66	64
Total Trp:Lys	20	19	18	19	18	19	18	18
CP, %	23.9	23.6	23.4	23.7	23.5	23.6	23.4	21.4
ME kcal/kg	3,387	3,408	3,428	3,385	3,385	3,394	3,401	3,410
Ca, %	0.88	0.88	0.88	0.89	0.89	0.88	0.88	0.80
P, %	0.80	0.78	0.77	0.80	0.80	0.79	0.78	0.75
Available P, %	0.47	0.47	0.47	0.47	0.47	0.47	0.47	0.42

¹Pigs were a common diet from weaning for 7 d, experimental diets were then fed for 14 d, followed by a 21 d common period.

²SBM = soybean meal; PepSoyGen (Nutra-Flo, Sioux City, IA).

³DPS 50 (Nutra-Flo, Sioux City, IA).

⁴Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B12; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

⁵Values for fish meal were from NRC (1998) and values for fermented soybean meal and dried porcine solubles were provided by the manufacturer.

Table 4. Effects of fish meal, fermented soybean meal, and dried porcine solubles on nursery pig performance (Exp. 1)¹

Item;	Control	Fish meal 5.00%	Dried porcine solubles ² 3.50%	Fermented SBM ³ 6.00%	Fermented SBM ³ + dried porcine solubles ² 1.75% + 1.75%	Fermented SBM ³ + fish meal 3.00% + 2.50%	SE
d 0 to 14							
ADG, g	252 ^a	268 ^a	313 ^b	269 ^a	302 ^b	255 ^a	14.6
ADFI, g	331 ^x	356 ^{xy}	366 ^y	343 ^{xy}	355 ^{xy}	334 ^x	18.2
G:F	0.75 ^{ax}	0.75 ^{ax}	0.86 ^{cy}	0.79 ^{abx}	0.85 ^{bcy}	0.76 ^{ax}	0.030
d 14 to 28							
ADG, g	516 ^{abxy}	535 ^{aby}	529 ^{abxy}	546 ^{by}	500 ^{ax}	540 ^{by}	18.0
ADFI, g	720 ^{xy}	724 ^{xy}	723 ^{xy}	740 ^{xy}	701 ^x	746 ^y	24.0
G:F	0.72	0.74	0.73	0.74	0.72	0.72	0.018
d 0 to 28							
ADG, g	383 ^{ay}	402 ^{abxy}	421 ^{bx}	407 ^{abx}	401 ^{abxy}	396 ^{aby}	13.4
ADFI, g	525	540	544	541	528	538	15.7
G:F	0.73 ^{bx}	0.74 ^{abxy}	0.77 ^{az}	0.75 ^{abxyz}	0.76 ^{abyz}	0.74 ^{bxy}	0.017

^{ab} Within a row, means without a common superscript differ ($P < 0.05$).

^{xyz} Within a row, means without a common superscript differ ($P < 0.10$).

¹A total of 252 pigs (initial BW 6.8 ± 1.8 kg) were used in a 14-d growth assay. Pigs were blocked by initial BW and randomly allotted to 1 of 6 treatments with 6 pigs/pen and 7 pens/treatment.

²DPS 50 (Nutra-Flo, Sioux City, IA).

³SBM = soybean meal; PepSoyGen (Nutra-Flo, Sioux City, IA).

Table 5. Effects of fish meal, fermented soybean meal, and the combination of fermented soybean meal and dried porcine solubles on nursery pig performance (Exp. 2)¹

Table 5: Effects of fish meal, fermented soybean meal, and the combination of fermented soybean meal and dried porcine solids on nursery pig performance (Exp. 2)														
Item	Control	P-value ²												
		Fish meal		Fermented SBM ³		Combination ⁴		SE	Fish meal		Fermented SBM		Combination	
		3.00%	6.00%	3.75%	7.50%	1.88% + 1.88%	3.75% + 3.75%		Lin	Quad	Lin	Quad	Lin	Quad
D 0 to 14														
ADG, ^{5,6} g	262	285	256	260	262	293	295	18.9	0.71	0.08	1.00	0.89	0.06	0.32
ADFI, ⁷ g	345	360	314	315	330	351	352	22.3	0.08	0.05	0.39	0.14	0.69	0.90
G:F ⁸	0.75	0.79	0.80	0.83	0.79	0.84	0.84	0.018	0.07	0.44	0.17	0.01	0.002	0.06
D 14 to 35														
ADG, g	587	596	580	579	592	572	597	24.9	0.74	0.45	0.77	0.52	0.60	0.24
ADFI, g	868	880	850	837	874	857	876	37.4	0.52	0.38	0.83	0.17	0.77	0.53
G:F	0.68	0.68	0.68	0.69	0.68	0.67	0.68	0.009	0.67	0.77	0.88	0.22	0.74	0.19
D 0 to 35														
ADG, g	455	472	446	451	458	460	476	22.3	0.58	0.17	0.91	0.73	0.24	0.70
ADFI, g	657	672	629	629	653	653	667	31.4	0.22	0.14	0.86	0.19	0.67	0.67
G:F	0.69	0.70	0.71	0.72	0.70	0.70	0.71	0.008	0.25	0.85	0.49	0.03	0.06	0.85

¹A total of 350 pigs (initial BW 6.1 ± 2.2 kg) were used in a 14-d growth assay. Pigs were blocked by initial BW and randomly allotted to 1 of 7 treatments with 5 pigs/pen and 10 pens/treatment.

²Linear (Lin) and quadratic (Quad) contrasts.

³SBM = soybean meal; PepSoyGen (Nutra-Flo, Sioux City, IA).

⁴Contained a 50:50 combination of fermented SBM and dried porcine solubles (DPS 50; Nutra-Flo, Sioux City, IA).

⁵Contrast: mean of fish meal vs. mean of combination, $P = 0.05$.

⁶Contrast: mean of fermented SBM vs. mean of combination, $P = 0.01$.

⁷Contrast: mean of fermented SBM vs. mean of combination, $P = 0.02$.

⁸Contrast: mean of fish meal vs. mean of combination, $P = 0.03$.

CHAPTER 2 - Efficacy of commercial enzymes in diets containing a variety of levels and sources of dried distillers grains with solubles for nursery pigs

Abstract

In 2 experiments, 530 pigs were used to evaluate the effects of adding of commercial enzymes to diets containing dried distillers grains with solubles (DDGS) on pig growth performance. In Exp. 1, 180 pigs (initial BW 9.0 kg) were fed either a corn–soybean meal-based control diet, a diet containing 30% corn DDGS, or the 30% DDGS diet with 0.05% of enzyme A, B, or C. There were 6 pigs per pen and 6 pens per treatment. Overall (d 0 to 27), neither DDGS nor enzyme addition influenced ($P > 0.10$) ADG and G:F. In Exp. 2, 350 pigs (initial BW 11.0 kg) were fed 1 of 10 dietary treatments. Pigs were fed either a control corn–soybean meal-based diet or the control diet containing 15% or 30% DDGS from 3 sources (corn, sorghum 1, or sorghum 2). Diets containing 30% DDGS were fed with or without enzyme A from Exp. 1. There were 5 pigs per pen and 7 pens per treatment. Overall (d 0 to 21), there were no ($P > 0.32$) enzyme \times DDGS source interactions observed. Corn DDGS did not influence pig performance ($P > 0.36$). Sorghum DDGS reduced ($P = 0.003$) G:F, with no difference between sorghum DDGS sources. Adding the commercial enzyme to the 30% DDGS diets did not improve ($P > 0.57$) performance. In summary, feeding diets with sorghum DDGS resulted in poorer G:F with no change in ADG compared with feeding the control diet or diets containing corn DDGS. Adding the enzymes used in this study to corn–soybean meal-based diets containing 30% DDGS did not improve growth performance.

Key words: carbohydrase, DDGS, distillers, enzyme, growth, pig

Introduction

Increased ethanol production has prompted the swine industry to increase its use of biofuel coproducts. Dried distillers grains with solubles (**DDGS**) is one such coproduct that is

widely used (Stein and Shurson, 2008). Studies show that 30% DDGS can replace the cereal grain source in nursery pig diets without affecting growth performance (Gaines et al., 2006; Spencer et al., 2007; Burkey et al., 2008). Little information on the impact of feeding sorghum-based DDGS is available in the nursery phase. Variability in DDGS nutrient content, specifically in Lys concentration and digestibility, has been reported (Stein and Shurson, 2008). One source of this variability is likely due to the variety of carbohydrate sources used in ethanol production (corn, sorghum, or wheat).

Because the majority of the starch fraction is removed by fermentation, other components, such as fiber, increase in concentration. This fiber fraction contains non-starch polysaccharides that the pig is unable to digest because of its lack of specific digestive enzymes. Supplemental enzymes have been developed for use in swine diets to assist in digestion of non-starch polysaccharides. These enzymes have been successful in increasing the digestibility of European swine diets, which are typically formulated with starch sources that have a high crude fiber component, such as barley, wheat, or rye (Omogbenigun et al., 2004). These commercial enzymes, such as various carbohydrases, are used to improve feed utilization and decrease the cost of gain (Partridge, 2001). Because corn is highly digestible and has a low fiber content, enzymes have not consistently shown improvements in growth performance when used in corn-based diets (Kim et al., 2003). Therefore, we speculate that enzymes may be more beneficial in diets containing DDGS than in corn–soybean meal-based diets. The objective of these experiments was to evaluate the effects of different commercial enzymes in diets containing a variety of sources of DDGS on nursery pig growth performance.

Materials and methods

General

All experimental procedures were approved by the Kansas State University Institutional Animal Care and Use Committee (number 2461).

Experiment 1

A total of 180 pigs (TR4 × 1050, PIC Hendersonville, TN; initial BW 9.0 kg) were used in a 27-d growth trial to evaluate the effects of 3 different commercial enzymes in diets

containing corn DDGS on nursery pig performance. Pigs were blocked by weight and allotted to 1 of 5 dietary treatments. There were 6 pigs per pen and 6 pens per treatment. Each pen (1.2 m²) contained 1 self-feeder and 1 nipple waterer to provide ad libitum access to feed and water. Pigs were housed in the Kansas State University Swine Teaching and Research Center in Manhattan.

A common pelleted starter diet was fed from weaning until the start of the experiment (d 7). The 5 dietary treatments fed were: (1) positive control, corn–soybean meal diet, (2) negative control, corn–soybean meal diet with 30% corn DDGS (Chief Ethanol Fuels, Hasting, NE), and the negative control diet with either (3) 0.05% enzyme A (Easyzyme Mixer 1; Archer Daniels Midland Company, Decatur, IL), (4) 0.05% enzyme B (Hemicell-2; Form-A-Feed, Inc, Stewart, MN), or (5) 0.05% enzyme C (Porzyme 93010; Danisco, New Century, MO; Table 1). Inclusion levels were chosen on the basis of manufacturers' recommendations and guaranteed analysis. Published mean standardized ileal digestible (SID) values for corn DDGS were used in diet formulation (Stein et al., 2006). Also, the ME value of the DDGS grains were used in diet formulation (3,420 kcal of ME/kg for corn and 3,340 kcal of ME/kg for sorghum; NRC, 1998). Treatment diets were fed for 27 d and were in meal form. Average daily gain, ADFI, and G:F were determined by weighing pigs and measuring feed disappearance on d 7, 14, and 27 of the trial.

Experiment 2

A total of 350 pigs (PIC C22 × 1050, PIC Hendersonville, TN; initial BW 11.0 kg) were used in a 21-d growth trial to evaluate the effects of a commercial enzyme in diets containing corn or sorghum DDGS on nursery pig performance. Pigs were blocked by weight and allotted to 1 of 10 dietary treatments. There were 5 pigs per pen and 7 pens per treatment. Each pen (1.5 m²) contained a 4-hole dry self-feeder and 1 cup waterer to provide ad libitum access to feed and water. The study was conducted at the Kansas State University Segregated Early Weaning Facility in Manhattan.

The 10 experimental treatments were: (1) positive control, corn–soybean meal diet, (2) 15% corn DDGS (Chief Ethanol Fuels, Hastings, NE), (3) 30% corn DDGS, (4) 30% corn DDGS + 0.05% enzyme A, (5) 15% sorghum DDGS from source 1 (Kansas Ethanol, Lyons, KS), (6) 30% sorghum DDGS from source 1, (7) 30% sorghum DDGS from source 1 + 0.05%

enzyme A, (8) 15% sorghum DDGS from source 2 (U.S. Energy Partners, Russell, KS), (9) 30% sorghum DDGS from source 2, and (10) 30% sorghum DDGS from source 2 + 0.05% enzyme A (Table 2). Sources of DDGS were analyzed for proximate analysis and amino acid levels (AOAC, 2000). Analyzed total AA levels for DDGS sources were used to calculate SID amino acid levels, which were used in diet formulation (Stein et al., 2006; Tables 3 and 4). Treatment diets were fed for 21 d. All diets were in meal form. Average daily gain, ADFI, and G:F were determined by weighing pigs and measuring feed disappearance on d 7, 14, and 21 of the trial.

Statistical Analysis

Data were analyzed as a completely randomized design with pen as the experimental unit. Data were analyzed by ANOVA with the MIXED procedure of SAS with treatment as a fixed effect. All possible pairwise comparisons were used to evaluate differences among treatments in Exp 1. Differences in Exp 2 were evaluated using preplanned contrasts. Linear and quadratic effects of increasing level of corn or sorghum DDGS using linear and quadratic effects with the corn-soy control treatment used as the first dosage level in both comparisons. One contrast compared all treatments containing corn DDGS to all treatments that contained Sorghum DDGS to evaluate the corn versus sorghum DDGS effect. One contrast compared all treatments containing sorghum DDGS from source 1 versus all treatments containing sorghum DDGS from source 2 to evaluate the effect of sorghum source. One contrast compared all treatments containing 30% DDGS without enzyme to all treatments containing 30% DDGS with enzyme to evaluate the effect of enzyme. Finally, one contrast was used to compare treatments containing enzyme to their respective DDGS source to determine an enzyme \times DDGS source interaction. Means were considered significant if their *P*-values were < 0.05 and trends if their *P*-values were < 0.10 .

Results

Chemical analysis

Corn DDGS had lower CP and fiber content than both sources of sorghum DDGS. Crude fat content of corn DDGS lower than that of sorghum DDGS from source 1, while similar to that of sorghum DDGS from source 2 (Table 3). Sorghum DDGS from source 1 had higher CP, fat,

fiber, and ash contents than sorghum DDGS from source 2. Levels of Lys and Trp were higher in corn DDGS than sorghum DDGS. Cys, Ile, Leu, Thr, and Val levels were all lower in corn DDGS than sorghum DDGS.

Analyzed levels of enzymes were all higher than formulated levels (Table 4). Proximate analysis of experimental diets showed similar nutrient values as formulated (Tables 5 and 6).

Experiment 1

Overall (d 0 to 27), pigs fed the positive and negative control diets had greater ($P < 0.05$) ADG and tended to have greater ($P < 0.10$) ADFI than pigs fed diets containing enzyme B (Table 7). Also, pigs fed the positive control diet had greater ($P < 0.05$) ADG than pigs fed diets containing enzyme A. There were no overall differences ($P > 0.10$) in G:F between treatment diets.

Experiment 2

Overall (d 0 to 21), there were no ($P > 0.32$) sorghum source or enzyme \times DDGS source interactions observed (Tables 8 and 9). Increasing dietary corn DDGS did not affect ($P > 0.36$) performance. Increasing dietary sorghum DDGS did not affect ($P > 0.11$) ADG or ADFI but decreased (linear, $P = 0.003$) G:F. Feeding sorghum DDGS rather than corn DDGS did not affect ($P = 0.76$) ADG but tended to decrease ($P = 0.06$) ADFI and decreased ($P = 0.05$) G:F. Enzyme addition did not influence ($P > 0.57$) ADG, ADFI, or G:F.

Discussion

Our data agrees with previous research showing 30% corn DDGS can be used in nursery pig diets (Gaines et al., 2006). Also, our data shows pigs fed increasing sorghum DDGS had decreased G:F, which is in agreement with data published by Feoli et al. (2008), who showed that inclusion of 30% sorghum DDGS lowered weanling pig ADG and G:F compared with a control diet without DDGS (670 vs. 548 g/d ADG and 689 vs. 585 g/kg G:F, respectively). Variability likely plays a role in the growth response to dietary DDGS. Stein and Shurson (2008) reported that the Lys content and digestibility in DDGS can vary greatly. As expected, pigs fed corn DDGS had improved G:F compared with those fed sorghum DDGS, likely because of a difference in energy between the diets. The energy value of corn is 3,420 kcal of ME/kg,

whereas that of sorghum is 3,340 kcal of ME/kg (NRC 1998). Thus, we would expect sorghum DDGS to have a lower energy content than that of corn DDGS.

All enzyme levels contained higher levels of dietary enzymes than the manufacturers guaranteed. Addition of these commercial enzymes to diets has been shown to improve performance in poultry (Bedford, 2000; Acamovic, 2001; Cowieson and Adeola, 2005), and these enzymes are widely used in pig diets in Europe, where feedstuffs with high fiber concentrations are the primary source of carbohydrates. Many cereal grains used in Europe have a high proportion of non-starch polysaccharides such as (1-3), (1-4)- β -D-glucans (barley and oats) and arabinoxylans (wheat, rye, and triticale; Bach Knudsen, 1997). Soybean meal, however, is high in β -galactomannans and α -1,6-galactosides, even after processing (Hartwig et al., 1997; Rackis, 1981). Carbohydrases such as α -galactosidase, β -1,4-mannanase, β -glucanase, and xylanase help break down some these insoluble bonds that the monogastric animal is otherwise unable to digest (Sugimoto and Van Buren, 1970; McGhee et al., 1978). Because corn is highly digestible and low in fiber, enzymes have not consistently shown improvements in growth performance (Partridge, 2001). Likewise, these experiments show that the enzymes tested did not improve weanling pig performance when added to diets containing 30 % DDGS compared with a corn–soybean meal positive control diet.

Because the distillation process increases the proportion of non-starch polysaccharides in cereal grains, it is theorized that dietary enzymes may be more effective when DDGS is included in the diet. However, both of our studies show that adding these enzymes had no effect on nursery pig performance whether dietary DDGS was fed or not. However, much is unknown regarding the mode of action of carbohydrases (Bedford, 2002). The effect of breaking these bonds seems to differ between poultry and swine. In poultry, carbohydrases appear to affect the viscosity of dietary ingredients within the gastrointestinal tract (GIT). Alimrall et al. (1995) found that increasing digesta viscosity in the GIT with carbohydrases may increase nutrient retention and utilization in high-fiber diets in poultry. However, Johansen et al. (1997) and Lindberg et al. (2003) suggested that altering GIT viscosity in swine by including carbohydrases does not result in nutrient utilization differences by pigs, because the majority of β -glucans are broken down prior to the terminal ileum. This is similar to the conclusions of Thacker et al.

(1992), who suggested that an increase in the digestibility of nutrients by swine is due to an increase in enzymatic degradation, not by a change in GIT viscosity.

The increase in enzymatic degradation, such as that of β -glucans, may allow the pig to utilize the sugars from non-starch polysaccharides for energy (Kim et al., 2003). This may be the reason for increased performance seen in some studies from adding carbohydrases. Also, dietary carbohydrases increase the sugar release in a diet and thus also increase lactobacilli growth (Axelsson, 1998). Höberg and Lindberg (2004) showed that supplementing high-fiber diets with fiber-degrading enzymes shifted the bacteria that dominate the ileum from acetic acid to lactic acid. Therefore, the increase in lactobacilli activity rather than gastrointestinal viscosity, may be the reason some pigs experience an increase in digestibility when fed carbohydrases; lactobacilli increases gut health and helps exclude pathogens (Pluske et al., 2001).

No peer-reviewed research shows the effects of the carbohydrases used in these experiments on nursery pig growth performance. Research involving a less concentrated form of Enzyme A (Porzyme 9300) shows that the enzyme increases the Ca and apparent AA digestibilities but does not affect growth performance of pigs fed wheat-based diets (Mavromichalis et al., 2000; Nortey et al., 2008; Woyengo et al., 2008).

Because of the variability in response to dietary carbohydrases on pig performance, it is difficult to explain the various responses, or lack of responses, from study to study. Our data suggest that the enzymes evaluated, when included at manufacturer suggested levels, did not improve growth performance. Also, adding different enzymes to diets containing 30% DDGS did not improve performance in a corn–soybean meal-based diet or a corn–soybean meal-based diet with 30% added DDGS. Feeding diets including sorghum DDGS resulted in poorer feed efficiency because of the lower energy content of sorghum DDGS.

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Figures and Tables

Table 1. Diet composition (Exp. 1, as-fed basis)¹

Item	Enzyme:	Corn-soy No	30% Corn DDGS ² Yes ³
Ingredient, %			
Corn		65.42	41.35
Soybean meal, 46.5% CP		30.55	25.05
Corn DDGS ²		---	30.00
Monocalcium P, 21% P		1.65	0.90
Limestone		0.98	1.38
Salt		0.35	0.35
Vitamin premix ⁴		0.25	0.25
Trace mineral premix ⁴		0.15	0.15
L-Lys HCl		0.37	0.45
DL-Met		0.15	0.06
L-Thr		0.14	0.06
Total		100.00	100.00
Calculated composition			
Total Lys, %		1.38	1.48
Standardized ileal digestibility, %			
Lys		1.25	1.25
Met:Lys		35	33
Met & Cys:Lys		59	59
Thr:Lys		63	63
Trp:Lys		17	17
Val:Lys		67	78
Lys:ME ratio, g/Mcal		3.79	4.12
CP, %		20.3	25.0
ME, kcal/kg		3,298	3,038
Ca, %		0.80	0.80
P, %		0.74	0.71
Available P, %		0.42	0.42

¹Pigs were fed experimental diets from d 0 to 21 of the trial.

²Dried distillers grains with solubles (DDGS).

³0.05% of Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL), Hemicell-W (Form-A-Feed, Inc., Stewart, MN), or Porzyme 93010 (Danisco Animal Nutrition, Marlborough, UK) were added to the 30% DDGS diet in place of corn.

⁴Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B₁₂; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄·H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

Table 2. Diet composition (Exp. 2, as-fed basis)¹

		Corn-soy	Corn DDGS ²		Source 1 Sorghum DDGS ²		Source 2 Sorghum DDGS ²	
		Control	15%	30%	15%	30%	15%	30%
Item	Enzyme:	No	No	Yes ³	No	Yes ³	No	Yes ³
Ingredient, %								
Corn		65.73	55.86	44.10	55.26	43.17	55.13	42.61
Soybean meal, 46.5% CP		30.24	25.38	22.55	25.88	23.22	25.97	23.73
DDGS ²		-	15.00	30.00	15.00	30.00	15.00	30.00
Monocalcium P, 21% P		1.63	1.25	0.85	1.30	0.95	1.30	1.00
Limestone		1.00	1.08	1.15	1.18	1.38	1.18	1.33
Salt		0.35	0.35	0.35	0.35	0.35	0.35	0.35
Vitamin premix ⁴		0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace mineral premix ⁴		0.15	0.15	0.15	0.15	0.15	0.15	0.15
L-Lys HCl		0.38	0.44	0.44	0.44	0.45	0.45	0.46
DL-Met		0.14	0.11	0.06	0.09	0.02	0.10	0.05
L-Thr		0.14	0.14	0.11	0.12	0.07	0.12	0.08
Total		100.00	100.00	100.00	100.00	100.00	100.00	100.00
Calculated composition								
Total Lys, %		1.38	1.42	1.47	1.41	1.45	1.41	1.44
Standardized ileal digestibility, %								
Lys		1.25	1.25	1.25	1.25	1.25	1.25	1.25
Met:Lys		34	33	31	32	29	32	30
Met & Cys:Lys		58	58	58	58	58	58	58
Thr:Lys		62	62	62	62	62	62	62
Trp:Lys		17	16	16	16	16	16	16
Val:Lys		66	67	67	71	79	71	78
Lys:ME ratio, g/Mcal		3.79	3.77	3.76	3.80	3.80	3.80	3.80
CP, %		20.2	21.0	22.5	21.9	24.2	21.8	24.1
ME kcal/kg		3,298	3,311	3,322	3,294	3,287	3,294	3,287
Ca, %		0.80	0.80	0.80	0.80	0.80	0.80	0.80
P, %		0.73	0.72	0.70	0.71	0.70	0.71	0.70
Available P, %		0.42	0.42	0.42	0.42	0.42	0.42	0.42

¹Pigs were fed experimental diets from d 0 to 21 of the trial.²Dried distillers grains with solubles (DDGS).³0.05% of Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL) was added to the 30% DDGS diet in place of corn.⁴Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B₁₂; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄·H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

Table 3. Chemical analysis of dried distillers grains with solubles (DDGS; Exp. 2, as-fed basis)¹

Item;	Corn DDGS	Source 1 Sorghum DDGS	Source 2 Sorghum DDGS
DM, %	88.50	88.34	88.43
Ash, %	5.13	4.06	3.91
Crude fat, %	8.93	10.22	8.91
Crude fiber, %	5.72	7.21	6.90
CP, %	25.94	30.74	29.67
Total amino acid, ² %			0.58
Cys	0.47	0.58	0.52
Ile	0.97	1.25	1.25
Leu	2.82	3.93	3.92
Lys	1.08	0.95	0.84
Met	0.52	0.58	0.50
Thr	0.97	1.09	1.03
Trp	0.22	0.20	0.19
Val	1.27	1.67	1.62

¹Results of analyzed values on which the diets were formulated.

²Total amino acid levels were multiplied times standardized ileal amino acid digestibilities (Stein et al, 2006) to calculate standardized ileal digestibility values used in diet formulation.

Table 4. Composition of enzymes (Exp. 1 & 2)

Item;	Enzyme A ¹		Enzyme B ²		Enzyme C ³	
	Guaranteed ⁴	Analyzed	Guaranteed ⁴	Analyzed	Guaranteed ⁴	Analyzed
α -Galactosidase, units/g	7	8.6	-	-	-	-
Galactomannanase, units/g	22	65.6	140,000	194,000	-	-
β -Gluconase, units/g	220	740.4	-	-	-	-
Xylanase, units/g	330	788.1	70,000	116,000	40,000	> 40,000

¹Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL). One unit is micromoles total reducing sugars released per minute at 30°C and pH 4.0.

²Hemicell-W (Form-A-Feed, Inc., Stewart, MN).

³Porzyme 93010 (Danisco Animal Nutrition, Marlborough, UK).

⁴Values provided by the manufacturer.

Table 5. Chemical analysis of diets (Exp. 1, as-fed basis)

Enzyme:	Corn-soy	30% Corn DDGS			
	None	None	Enzyme A ¹	Enzyme B ²	Enzyme C ³
DM, %	87.07	88.71	88.86	88.42	88.75
CP, %	19.88	22.56	24.06	22.97	24.15
Crude fat, %	2.18	4.54	4.43	4.26	4.64
Crude fiber, %	2.24	3.70	3.73	3.57	3.69
Ash, %	5.67	7.08	7.24	6.80	7.18

¹Easzyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL).

²Hemicell-W (Form-A-Feed, Inc., Stewart, MN).

³Porzyme 93010 (Danisco Animal Nutrition, Marlborough, UK).

Table 6. Chemical analysis of diets (Exp. 2, as-fed basis)

Enzyme ¹ :	Control	Corn DDGS			Source 1 Sorghum DDGS			Source 2 Sorghum DDGS		
		15%	30%	30%	15%	30%	30%	15%	30%	30%
	No	No	No	Yes	No	No	Yes	No	No	Yes
DM, %	86.98	87.60	88.28	88.48	87.64	88.62	87.48	87.08	87.81	87.50
CP, %	21.08	20.51	23.07	22.94	20.82	23.57	24.10	20.90	23.00	23.10
Crude fat, %	3.15	4.39	5.21	4.75	3.60	4.68	4.36	3.14	4.14	4.10
Crude fiber, %	2.37	2.84	3.27	3.40	3.01	3.90	3.55	2.71	3.76	3.45
Ash, %	5.49	5.44	5.81	5.66	5.55	5.40	5.71	5.32	5.29	5.10

¹Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL).

Table 7. Effects of dried distillers grains and enzymes on nursery pig performance (Exp. 1)¹

Item;	No Enzyme	30% DDGS				SE
		No Enzyme	Enzyme A ²	Enzyme B ³	Enzyme C ⁴	
d 0 to 27						
ADG, g	531 ^a	512 ^{ab}	500 ^{bc}	475 ^c	521 ^{ab}	14.9
ADFI, g	769 ^{abx}	772 ^{abx}	728 ^{abxy}	701 ^{by}	789 ^{ax}	37.9
G:F	0.69	0.67	0.69	0.68	0.66	0.022

¹A total of 180 pigs (6 pigs per pen and 6 pens per treatment) with an initial BW of 9.0 kg. Pigs were fed a common diet from weaning until the start of the trial, then fed experimental diets for 27 d.

²Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL).

³Hemicell-W (Form-A-Feed, Inc., Stewart, MN).

⁴Porzyme 93010 (Danisco, New Century, MO).

^{abc}Within a row, means without a common superscript differ ($P < 0.05$).

^{xy}Within a row, means without a common superscript differ ($P < 0.10$).

Table 8. Effects of dried distillers grains with solubles (DDGS) and enzymes on nursery pig performance (Exp. 2)¹

Enzyme ² :	Corn-soy	Corn DDGS			Source 1 Sorghum DDGS			Source 2 Sorghum DDGS		
	Control	15%	30%	30%	15%	30%	30%	15%	30%	30%
	No	No	No	Yes	No	No	Yes	No	No	Yes
d 0 to 21										
ADG, g	476	461	467	467	487	458	445	478	462	472
ADFI, g	727	725	726	734	761	747	713	762	763	765
G:F	0.66	0.64	0.64	0.64	0.64	0.61	0.62	0.63	0.61	0.62

¹Pigs were fed experimental diets from d 0 to 21 of the trial.

²Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL).

Table 9. Contrast *P*-values for the effects of dried distillers grains and enzymes on nursery pig performance (Exp. 2)¹

Probability, <i>P</i> <									
Item;	Corn DDGS		Sorghum DDGS		Corn DDGS vs. Sorghum DDGS	Sorghum 1 vs. Sorghum 2 DDGS	30% DDGS vs. 30% DDGS + Enzyme	Enzyme ² × DDGS Source	SEM
	Linear	Quadratic	Linear	Quadratic					
D 0 to 21									
ADG, g	0.53	0.37	0.19	0.12	0.76	0.40	0.87	0.91	10.0
ADFI, g	0.98	0.94	0.19	0.20	0.06	0.12	0.58	0.35	17.4
G:F	0.49	0.37	0.003	0.88	0.05	0.39	0.67	0.33	0.012

¹Pigs were fed experimental diets from d 0 to 21 of the trial.²Easyzyme Mixer 1 (Archer Daniels Midland Company, Decatur, IL).

CHAPTER 3 - An evaluation of Peptone as a specialty protein source in diets for nursery pigs

Abstract

Two experiments were conducted to evaluate the effects of select menhaden fish meal (SMFM), spray-dried animal plasma (SDAP), or two forms of a spray-dried ultra-filtrated porcine intestinal mucosa (Peptone) on nursery pig performance. Pigs were fed experimental phase 1 diets from d 0 to 10 post-weaning, followed by experimental phase 2 diets from d 10 to d 20 (Exp. 1) or d 25 (Exp. 2). Pigs were then fed a common phase 3 diet that contained no specialty proteins for 7 d. There were 6 replications per treatment and 6 pigs per pen in both experiments. In Exp. 1, 216 weanling pigs (PIC TR4 × 1050, initial BW 5.4 kg) were fed either (1) a control diet containing no specialty protein sources; or the control diet with (2) 4% SMFM during phase 1 and 2% SMFM during phase 2; (3) 4% SDAP during phase 1 and no specialty protein sources during phase 2; (4) 4% SDAP during phase 1 and 2% SDAP during phase 2; (5) 4% Peptone 1 during phase 1 and no specialty protein sources during phase 2; or (6) 4% Peptone 1 during phase 1 and 2% Peptone 1 during phase 2. From d 0 to 10, pigs fed different diets had similar ($P < 0.10$) ADG. Also, pigs fed the control diet or diets containing SMFM tended to have improved ($P < 0.10$) G:F compared to pigs fed diets containing Peptone 1. From d 10 to 20, pigs previously fed 4% Peptone 1 during phase 1 and the control diet during phase 2 had improved ($P < 0.05$) ADG and ADFI compared to pigs fed 2% SDAP. Overall (d 0 to 27), specialty protein source did not influence ($P > 0.10$) ADG, ADFI, or G:F. In Exp. 2, 180 weanling pigs (PIC C22 × 1050, initial BW 5.9 kg) were fed either a (1) control diet containing no specialty protein sources; or the control diet with (2) 4% SMFM during phase 1 and 2% SMFM during phase 2; (3) 4% SDAP during phase 1 and no specialty protein sources during phase 2; (4) 4% SDAP during phase 1 and 2% SDAP during phase 2; (5) 4% Peptone 2 during phase 1 and no specialty protein sources during phase 2; or (6) 4% Peptone 2 during phase 1 and 2% Peptone during phase 2. From d 0 to 10, pigs fed diets containing Peptone 2 tended to have improved ($P < 0.10$) G:F compared to pigs fed the control diet. From d 10 to 25, pigs fed 2% Peptone 2 or SMFM had improved ($P < 0.05$) G:F compared to pigs fed the control diet. Overall (d 0 to 32), pigs fed 4%

Peptone 2 during phase 1 and 2% Peptone 2 during phase 2 had improved ($P < 0.05$) ADG compared to pigs fed SMFM, and improved ($P < 0.05$) G:F compared to pigs fed all other diets. Therefore, the Peptone products evaluated in these studies can be used in nursery pig diets without negatively affecting pig growth performance.

Key Words: growth, nursery pig, protein source, spray-dried intestinal mucosa

Introduction

Weanling pig diets often contain animal protein sources, such as fish meal and spray-dried animal plasma (SDAP), which have desirable amino acid profiles. Fish meal contains amino acids that are often deficient in cereal grains, in addition to vitamins and minerals that are often deficient in other protein sources (Church and Kellems, 1998). Research has shown that dietary fish meal supplementation increases nursery pig growth performance and disease resistance (Kim and Easter, 2001; Young et al., 2002; and Gaines et al., 2005). Spray-dried animal plasma is widely used in diets immediately post-weaning as it has consistently shown to improve weanling pig performance during the first week post-weaning by improving feed intake (Kats et al., 1994; Maxwell and Carter, 2001). The mode of action of these specialty proteins for improving nursery pig performance is not fully understood, but may be due to appetite stimulation or immunoglobulin concentrations (Pierce et al., 2005).

Another possible protein source for nursery diets is Peptone, which is a product made by ultra-filtrating porcine intestinal mucosa. This filtration process removes the amino-acid rich peptides from the mucosa, which is then spray-dried. The resulting material contains a high level of digestible peptides and amino acids. Research has shown that newly weaned pigs have a considerable capacity to absorb peptides in the small intestine (Gilbert et al., 2008). Thereby, this newly-developed Peptone protein source may provide an alternative to other traditional animal protein sources in nursery diets. While there is no research data available for this new Peptone product, previous research with dried porcine intestinal mucosa have shown improvements in piglet growth performance (Zimmerman et al., 1997; Lindemann et al., 1998; Carter et al., 1999; DeRouchey et al., 2003). Therefore, the objective of these experiments was to evaluate the effects of fish meal, spray-dried animal plasma, and Peptone on growth performance of weanling pigs.

Materials and methods

General

The experimental protocols used in these studies were approved by the Kansas State University Institutional Animal Care and Use Committee (#2461). Both forms of Peptone (Peptone 1, Exp. 1; Peptone 2, Exp. 2) were analyzed for DM, CP, percentage ash, Ca, P, Na, Cl, S, and AA concentrations (AOAC, 2000; Table 1).

Experiment 1

A total of 216 weanling pigs (PIC TR4 \times 1050, initial BW 5.4 kg) were used in a 27-d growth trial. Pigs were blocked by weight and allotted to 1 of 6 diets. There were 6 pigs per pen and 6 pens per treatment. Each pen (1.2 m²) contained 1 self-feeder and 1 nipple waterer to provide *ad libitum* access to feed and water. Pigs were housed in the Kansas State University Swine Teaching and Research Center.

The 6 experimental diets were: (1) control diet containing no specialty protein sources; or the control diet with (2) 4% SMFM during phase 1 and 2% SMFM during phase 2; (3) 4% SDAP during phase 1 and no specialty protein sources during phase 2; (4) 4% SDAP during phase 1 and 2% SDAP during phase 2; (5) 4% Peptone 1 during phase 1 and no specialty protein sources during phase 2; or (6) 4% Peptone 1 during phase 1 and 2% Peptone 1 during phase 2 (Table 2). Phase 1 diets were fed from d 0 to 10, phase 2 diets were fed from 10 to 20 d, and then all pigs were fed a common diet without any specialty protein sources for 7 d. All diets were fed in meal form. Analyzed nutrient values from Peptone 1 were used in diet formulation. The analyzed values were very similar to those of SDAP (NRC, 1998), and because standardized ileal digestible (SID) values were not available on Peptone 1, diets were formulated with SDAP SID percentages (NRC, 1998). Pigs were weighed and feed disappearance was measured on d 0, 5, 10, 17, 20, and 27 of the trial to determine ADG, ADFI, and G:F.

Experiment 2

A total of 180 weanling pigs (PIC TR4 \times 1050, initial BW 5.9 kg) were used in a 32-d growth trial. Pigs were blocked by weight and allotted to 1 of 6 diets. There were 5 pigs per pen and 6 pens per treatment. Each pen (1.5 m²) contained 1 self-feeder and 1 nipple waterer to

provide *ad libitum* access to feed and water. Pigs were housed in the Kansas State University Segregated Early Weaning Facility.

The 6 experimental diets were: (1) control diet containing no specialty protein sources; or the control diet with (2) 4% SMFM during phase 1 and 2% SMFM during phase 2; (3) 4% SDAP during phase 1 and no specialty protein sources during phase 2; (4) 4% SDAP during phase 1 and 2% SDAP during phase 2; (5) 4% Peptone 2 during phase 1 and no specialty protein sources during phase 2; or (6) 4% Peptone 2 during phase 1 and 2% Peptone during phase 2. Phase 1 diets were fed from d 0 to 10, phase 2 diets were fed from 10 to 25 d, and then all pigs were fed a common diet without specialty protein sources for 7 d. Phase 1 and 2 diets were pelleted, while the common phase 3 diet was in meal form. Proximate analysis of Peptone 2 showed a similar CP level as Peptone 1, thus the analyzed nutrient values from Peptone 1 were used in diet formulation. Pigs were weighed and feed disappearance was measured on d 0, 10, 18, 25, and 32 of the trial to determine ADG, ADFI, and G:F.

Statistical Analysis

Data were analyzed as a randomized complete block design with pen as the experimental unit. Data were analyzed by using an ANOVA in the MIXED procedure of SAS with the weight block as a random effect and treatments as a fixed effect. All possible pairwise comparisons were used to evaluate differences among treatments in Exp. 1 and Exp. 2. Means were considered significant if their *P*-values were < 0.05 and trends if their *P*-values were < 0.10.

Results

Peptone Composition

Crude protein levels were similar between the two Peptones, but Peptone 2 had over twice as much Lys than Peptone 1. Peptone 2 also had greater Thr, Met, and Trp levels than Peptone 1. Peptone 2 contained 5 percentage units more moisture, and had higher crude fat, Na, and Cl concentrations than Peptone 1. Peptone 1 and 2 had similar, relatively high S levels at 4.7%.

Experiment 1

From d 0 to 5, pigs fed the diets containing Peptone 1 tended to have improved ($P < 0.10$) ADG and had greater ($P < 0.05$) ADFI compared to pigs fed the control diet. Pigs fed diets containing SMFM also tended to have greater ($P < 0.10$) ADFI compared to pigs fed the control diet. From d 5 to 10, pigs fed diets containing SDAP tended to have improved ($P < 0.10$) ADG compared to pigs fed diets containing Peptone 1, and greater ($P < 0.10$) ADFI compared to pigs fed the control diet. From d 0 to 10, pigs fed different diets had similar ($P < 0.10$) ADG. Also, pigs fed the control diet or diets containing SMFM tended to have improved ($P < 0.10$) G:F compared to pigs fed diets containing Peptone 1.

During phase 2 (d 10 to 20), pigs previously fed 4% Peptone 1 during phase 1 and the control diet during phase 2 had improved ($P < 0.05$) ADG and ADFI compared to pigs previously fed 4% SDAP during phase 1 and 2% SDAP during phase 2. Pigs previously fed 4% Peptone 1 during phase 1 and 2% Peptone 1 during phase 2 and pigs fed the control diet tended to have improved ($P < 0.10$) ADG compared to pigs previously fed 4% SDAP during phase 1 and 2% SDAP during phase 2. Pigs previously fed 4% Peptone 1 during phase 1 and 2% Peptone 1 during phase 2 tended to have improved ($P < 0.10$) G:F compared to pigs previously fed 4% SDAP during phase 1 and the control diet during phase 2.

During the common period (d 20 to 27), ADG was similar ($P > 0.54$) among pigs previously fed different diets. Pigs previously fed the control diet for phase 1 had greater ($P < 0.05$) ADFI compared to pigs previously fed 4% Peptone 1 during phase 1 and tended to have greater ($P < 0.10$) ADFI compared to pigs previously fed 5.75% SMFM during phase 1 and 2.88% SMFM during phase 2. Also, pigs previously fed 4% SDAP during phase 1 and the control diet during phase 2 tended to have improved ($P < 0.10$) ADFI compared to pigs previously fed 4% Peptone 1 during phase 1. Pigs previously fed diets containing 4% Peptone 1 during phase 1 had tended to have improved ($P < 0.10$) G:F compared to pigs previously fed 4% SDAP or the control diet during phase 1.

Overall (d 0 to 27), pigs fed the various diets had similar ($P > 0.10$) ADG, ADFI, and G:F.

Experiment 2

From d 0 to 5, pigs fed the various diets had wide numerical variation in ADG, ADFI, and G:F, but all were statistically similar ($P > 0.10$). From d 0 to 10, pigs fed diets containing Peptone 2 had improved ($P < 0.05$) G:F compared to pigs fed the control diet. During phase 2 (d 10 to 25), pigs fed the various diets had similar ($P > 0.14$) ADG and ADFI. Pigs previously fed diets containing 4% SMFM or Peptone 2 during phase 1 and 2% SMFM or Peptone 2 during phase 2 had improved ($P < 0.05$) G:F compared to pigs fed the control diet, and tended to have improved ($P < 0.10$) G:F compared to pigs previously fed 4% SDAP during phase 1 and 2% SDAP during P2. Pigs previously fed 4% Peptone 2 during phase 1 and 2% Peptone 2 during phase 2 also tended to have improved ($P < 0.10$) G:F compared to pigs previously fed 4% Peptone 2 during phase 1 and the control diet during phase 2.

During the common period (d 25 to 32), all pigs had similar ($P > 0.21$) ADG. Pigs previously fed 4% Peptone 2 during phase 1 and 2% Peptone 2 during phase 2 tended to have improved ($P < 0.10$) ADFI compared to pigs previously fed 4% SMFM during phase 1 and 2% SMFM during phase 2. Pigs previously fed the control diet or diets containing 4% Peptone 2 during phase 1 and 2% Peptone 2 during phase 2 had improved ($P < 0.05$) G:F, while pigs previously fed the control diet or 4% SDAP during phase 1 and 2% SDAP during phase 2 tended to have improved ($P < 0.10$) G:F compared to pigs previously fed 4% SMFM during phase 1 and 2% SMFM during phase 2.

Overall (d 0 to 32), pigs fed Peptone 2 during phase 1 and 2 had improved ($P < 0.05$) ADG compared to pigs fed SMFM during phase 1 and 2, and tended to have improved ($P < 0.10$) ADG compared to pigs fed the control diet. All pigs had similar ($P > 0.19$) ADFI. Finally, pigs fed Peptone 2 during phase 1 and 2 had improved ($P < 0.05$) G:F compared to pigs fed all other diets.

Discussion

Fish meal has been shown to increase growth performance and disease resistance in nursery pigs (Kim and Easter, 2001; Young et al., 2002; Gaines et al., 2001). However, the pigs fed SMFM in both of our experiments had similar overall growth performance as the control. Previous research suggests that pig health status may affect the level of growth response in pigs fed diets containing SMFM (Maxwell and Carter, 2001). In one study, disease-challenged pigs

had a greater response to supplemented fish meal than healthier pigs (Bergstrom et al., 1997). There were no disease challenges to the pigs in these experiments, which may help explain the lack of response to fish meal in these experiments.

While adding SMFM resulted in no added benefit to weanling pig diets in this study, feeding dietary SDAP yielded mixed effects. Little benefit was seen by adding SDAP in Exp. 1, similar to results seen by Lenehan et al. (2007). This lack of response in growth performance from the specialty protein may be because, again, the pigs were of high-health. Research suggests that, similar to fish meal, pigs fed SDAP respond more favorably to the specialty protein when they are in a more challenging environment (Coffey and Cromwell, 1995 and Bergstrom et al., 1997).

However, improvements were seen in pig performance with SDAP supplementation in Exp. 2, even though pigs were of high-health. This is in agreement with previous research that has shown improvements in growth performance by feeding weanling pig diets with SDAP (Coffey and Cromwell, 1995; and de Rodas et al., 1995). Generally, the improvements in pig growth performance are more prominent during the first week post-weaning, and there is no added benefit in feeding SDAP after one week post-weaning (van Dijk et al, 2001). We found a similar effect as there was an improvement in performance from adding SDAP from d 0 to 5 compared to the control, but there was no overall benefit at the end of the experiment. With the difficulties of stimulating intake during the first days post-weaning, however, the use of SDAP is important to improve post-weaning performance.

It is unknown why diets with the same formulation yielded two different responses to specialty protein sources from two different groups of pigs housed in similar environments. The only difference between the diets is that those in Exp. 1 were in meal form, while those in Exp. 2 were pelleted. More research is needed, but it appears that there may be a potential relationship between pelleting and level of response seen with SDAP supplementation.

Some differences in Peptone chemical analysis were expected because the two different forms of specialty protein were ultra-filtrated with different size filters. However, the amplitude of change in some AA values, such as Lys, was surprising given that the Peptones had similar CP levels. While there is no data showing the effects of Peptone on nursery pig growth performance, a similar protein product, dried porcine solubles, has shown consistent improvement in piglet growth performance (Zimmerman et al., 1997; Lindemann et al., 1998;

Carter et al. 1999; DeRouchey et al., 2003). Our data suggests that the Peptone used in our experiments mimic the positive effects of dried porcine solubles. However, more research is needed to directly compare these two specialty protein sources.

In conclusion, the Peptone products evaluated in these studies can be used in nursery pig diets as an effective animal protein source to improve growth performance.

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Figures and Tables

Table 1. Analyzed composition of Peptone (Exp. 1 and 2; as-fed basis)¹

Item	Peptone 1 ²	Peptone 2 ³
DM, %	96.60	91.23
CP, %	74.59	74.21
Crude fat, %	0.23	1.48
Ash, %	16.88	17.68
Ca, %	0.07	0.11
P, %	0.98	1.01
Na, %	5.33	6.57
Cl, %	0.42	2.88
S, %	4.67	4.69
Amino acids, %		
Arg	3.30	4.59
His	0.97	1.82
Ile	2.12	3.03
Leu	3.28	5.44
Lys	2.70	6.35
Met	0.62	1.02
Phe	1.35	2.46
Thr	1.99	3.01
Trp	0.33	0.44
Val	2.61	3.81
Ala	2.63	3.49
Cys	1.29	1.07
Gly	6.36	5.04
Orn	1.01	0.52
Pro	4.25	3.63
Ser	1.25	2.73
Tau	0.09	0.24
Tyr	1.07	2.54

¹Analyzed by the University of Missouri Agriculture Experiment Station Chemical Laboratories.

²Peptone 1 was used in Exp. 1. Analyzed nutrient values were used in diet formulation. Analyzed values were similar to that of spray-dried animal plasma; and because standardized ileal digestible values were not available on Peptone 1 at the time of formulation, diets were formulated with SID percentages for spray-dried animal plasma.

³Peptone 2 was used in Exp. 2. Analyzed amino acid values were unavailable at diet formulation. However, analyzed CP levels were similar to that of Peptone 1. Thus, diets were formulated with the same values as Peptone 1.

Table 2. Diet composition (Exp. 1 and 2; as-fed basis)¹

Item	Phase 1 ²				Phase 2 ³				Phase 3 ⁴
	Control	4% Fishmeal	4% SDAP ⁵	4% Peptone 1 ⁶	Control	2% Fishmeal	2% SDAP ⁵	2% Peptone 2 ⁶	Common
Ingredient, %									
Corn	40.08	46.58	46.10	45.67	57.23	60.45	60.25	60.05	61.18
Soybean meal, 46.5% CP	40.28	30.35	30.37	30.34	37.82	32.86	32.87	32.85	33.85
Spray-dried animal plasma	---	---	4.00	---	---	---	2.00	---	---
Peptone	---	---	---	4.00	---	---	---	2.00	---
Select menhaden fish meal	---	4.00	---	---	---	2.00	---	---	---
Spray dried whey	15.00	15.00	15.00	15.00	---	---	---	---	---
Soybean oil	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.00
Monocalcium phosphate, 21% P	0.93	0.45	0.70	0.83	1.15	0.90	1.00	1.08	1.65
Limestone	0.98	0.73	1.15	1.10	1.03	0.93	1.13	1.10	0.95
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.35
Vitamin premix ⁷	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace mineral premix ⁷	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
L-Lys·HCl	0.20	0.29	0.20	0.40	0.25	0.30	0.25	0.35	0.30
DL-Met	0.16	0.17	0.14	0.19	0.13	0.14	0.12	0.14	0.12
L-Thr	0.08	0.14	0.05	0.16	0.10	0.13	0.09	0.14	0.11
L-Val	---	---	---	0.02	---	---	---	---	---
Phytase ⁸	0.09	0.09	0.09	0.09	0.09	0.09	0.09	0.09	0.09
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Calculated composition ⁹									
Total Lys, %	1.61	1.60	1.60	1.59	1.49	1.48	1.48	1.48	1.42
Standardized ileal digestibilities, %									
Lys	1.45	1.45	1.45	1.45	1.34	1.34	1.34	1.34	1.25
Met:Lys	33	35	31	34	33	35	31	33	31
Met & Cys:Lys	58	58	58	58	58	59	58	58	57
Thr:Lys	63	63	63	63	63	63	63	63	64
Trp:Lys	19	17	19	16	19	18	19	17	18
CP, %	24.3	22.9	23.3	23.0	22.8	22.1	22.3	22.2	21.4
ME, kcal/kg	3,345	3,372	3,369	3,340	3,373	3,387	3,386	3,372	3,347
SID Lys:ME, g/Mcal	4.34	4.30	4.30	4.34	3.97	3.96	3.96	3.97	4.23
Ca, %	0.80	0.80	0.80	0.80	0.75	0.75	0.75	0.75	0.80
P, %	0.69	0.66	0.66	0.66	0.66	0.65	0.64	0.64	0.75
Available P, %	0.48	0.48	0.48	0.48	0.42	0.42	0.42	0.42	0.42

¹A total of 396 nursery pigs (initial BW 5.4 or 5.9 kg) were used in a 25 or 32-d growth assay to determine the effect of protein source on growth performance.

²Phase 1 diets were fed from d 0 to 10.

³Phase 2 diets were fed from d 10 to 20 (Exp. 1) or from d 10 to 25 (Exp. 2).

⁴Phase 3 diets were fed from d 20 to 27 (Exp. 1) or from d 25 to 32 (Exp. 2).

⁵Spray-dried animal plasma.

⁶Peptone (Form-A-Feed, Inc.; Stewart, MN).

⁷Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B₁₂; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄·H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

⁸NatuPhos 600 (BASF Animal Nutrition; Mount Olive, NJ) provided 509 FTU/kg of diet, with a release of 0.10 available P.

⁹Nutrient values for fish meal and SDAP were from NRC (1998).

Table 3. Effects of protein source on nursery pig performance¹ (Exp. 1)

Item;	Phase 1 ² :	Control	4% SMFM	4% SDAP		4% Peptone 1		SE
	Phase 2 ³ :	Control	2% SMFM	Control	2% SDAP	Control	2% Peptone 1	
d 0 to 5								
ADG, g		135 ^{bz}	159 ^{byz}	142 ^{byz}	132 ^{bz}	198 ^{ax}	169 ^{abxy}	14.7
ADFI, g		101 ^{abx}	133 ^{bcdyz}	122 ^{abxxy}	93.5 ^{ax}	142 ^{cdyz}	160 ^{dz}	13.3
G:F		1.37 ^{abx}	1.22 ^{abxy}	1.33 ^{abx}	1.43 ^{ax}	1.40 ^{ax}	1.06 ^{by}	0.128
d 5 to 10								
ADG, g		242 ^{abxyz}	254 ^{abxyz}	278 ^{abxy}	291 ^{ax}	218 ^{cz}	224 ^{bcz}	23.2
ADFI, g		278 ^{bz}	290 ^{abyz}	332 ^{abxy}	338 ^{ax}	323 ^{abxyz}	284 ^{abz}	22.6
G:F		0.87 ^{ax}	0.88 ^{ax}	0.84 ^{abx}	0.87 ^{ax}	0.68 ^{by}	0.79 ^{abxy}	0.055
d 0 to 10								
ADG, g		188	206	210	213	208	196	15.8
ADFI, g		188 ^{ax}	211 ^{abxy}	227 ^{aby}	216 ^{abxy}	231 ^{by}	222 ^{abxy}	15.4
G:F		1.00 ^{ax}	0.98 ^{ax}	0.93 ^{abxyz}	0.99 ^{ax}	0.90 ^{abyz}	0.88 ^{bz}	0.043
d 10 to 20								
ADG, g		326 ^{abx}	304 ^{abxy}	296 ^{abxy}	279 ^{ay}	333 ^{bx}	332 ^{abx}	21.6
ADFI, g		431 ^{ab}	410 ^{ab}	408 ^{ab}	382 ^a	448 ^b	432 ^{ab}	25.3
G:F		0.75 ^{xy}	0.74 ^{xy}	0.73 ^x	0.73 ^{xy}	0.75 ^{xy}	0.77 ^y	0.022
d 20 to 27								
ADG, g		451	448	440	439	443	449	14.3
ADFI, g		944 ^{bz}	849 ^{abxy}	917 ^{abyz}	906 ^{abxyz}	828 ^{ax}	827 ^{ax}	36.1
G:F		0.49 ^x	0.53 ^{xy}	0.49 ^x	0.48 ^x	0.54 ^y	0.54 ^y	0.022
d 0 to 27								
ADG, g		301	303	300	295	314	312	14.1
ADFI, g		457	446	466	453	464	457	10.1
G:F		0.66	0.68	0.64	0.65	0.68	0.68	0.022

¹A total of 216 pigs (6 pigs/pen and 6 pens/treatment with an initial BW of 5.4 kg.²Pigs were fed phase 1 diets from d 0 to 10.³Pigs were fed phase 2 diets from d 10 to 20.^{ab}Within a row, means without a common superscript differ ($P < 0.05$).^{xyz}Within a row, means without a common superscript differ ($P < 0.10$).

Table 4. Effects of protein source on nursery pig performance¹ (Exp. 2)

Item;	Phase 1 ² :	Control	4% SMFM	4% SDAP		4% Peptone 2		SE
	Phase 2 ³ :	Control	2% SMFM	Control	2% SDAP	Control	2% Peptone 2	
d 0 to 5								
ADG, g		84 ^{cz}	93 ^{bcz}	107 ^{abcyz}	151 ^{ax}	100 ^{bcz}	137 ^{abxy}	16.8
ADFI, g		87 ^{byz}	84 ^{bz}	100 ^{byz}	131 ^{ax}	89 ^{byz}	104 ^{by}	13.3
G:F		0.89 ^{by}	1.14 ^{abxy}	1.01 ^{aby}	1.15 ^{abxy}	1.11 ^{abxy}	1.33 ^{ax}	0.135
d 5 to 10								
ADG, g		270	270	266	289	274	280	18.7
ADFI, g		305	293	304	324	287	290	16.5
G:F		0.90	0.92	0.88	0.89	0.96	0.96	0.046
d 0 to 10								
ADG, g		177 ^{bz}	181 ^{byz}	187 ^{abyz}	220 ^{ax}	185 ^{abyz}	209 ^{abxy}	12.6
ADFI, g		196 ^{by}	189 ^{by}	202 ^{aby}	227 ^{ax}	186 ^{by}	197 ^{by}	10.2
G:F		0.91 ^{bz}	0.96 ^{abyz}	0.92 ^{byz}	0.97 ^{abyz}	1.00 ^{abxy}	1.06 ^{ax}	0.037
d 10 to 25								
ADG, g		440	450	435	449	452	463	15.1
ADFI, g		627	603	605	634	639	616	21.8
G:F		0.70 ^{bz}	0.75 ^{axy}	0.72 ^{abxyz}	0.71 ^{abz}	0.71 ^{abyz}	0.75 ^{ax}	0.015
d 25 to 32								
ADG, g		599	557	603	595	610	629	31.5
ADFI, g		907 ^{xy}	909 ^y	938 ^{xy}	908 ^{xy}	962 ^{xy}	937 ^x	34.7
G:F		0.66 ^{ax}	0.61 ^{by}	0.64 ^{abxy}	0.65 ^{abx}	0.63 ^{abxy}	0.67 ^{ax}	0.017
d 0 to 32								
ADG, g		393 ^{aby}	389 ^{by}	402 ^{abxy}	399 ^{abxy}	402 ^{abxy}	420 ^{ax}	13.0
ADFI, g		554	541	555	559	567	555	17.0
G:F		0.71 ^b	0.72 ^b	0.72 ^b	0.71 ^b	0.71 ^b	0.76 ^a	0.010

¹A total of 180 pigs (6 pigs/pen and 6 pens/treatment with an initial BW of 5.9 kg.²Pigs were fed phase 1 diets from d 0 to 10.³Pigs were fed phase 2 diets from d 10 to 25.^{ab}Within a row, means without a common superscript differ ($P < 0.05$).^{xyz}Within a row, means without a common superscript differ ($P < 0.10$).

CHAPTER 4 - Efficacy of different commercial phytase enzymes and development of an available phosphorus release curve

Abstract

In 2 experiments, 184 pigs (PIC, 10.3 and 9.7 kg BW, respectively) were used to develop an available P (aP) release curve for commercial phytase products. Pigs in both experiments were fed a basal diet (0.06% aP) and levels of added aP from inorganic P to develop a standard curve. In Exp. 1, 100, 175, 250, or 500 phytase units (FTU)/kg OptiPhos 2000 – M or 200, 350, 500, or 1,000 FTU/kg Phyzyme XP were added to the basal diet. In Exp. 2, 250, 500, 750, or 1,000 FTU/kg OptiPhos 2000 – M; 500, 1,000, or 1,500 FTU/kg Phyzyme XP; or 1,850 or 3,700 phytase units (FYT)/kg Ronozyme – P (M) were added to the basal diet. Manufacturer-guaranteed phytase levels were used in diet formulation. Diets were analyzed for phytase with both the Phytex and AOAC methods. Pigs were blocked by sex and weight and allotted to individual pens with 8 pens per treatment. Pigs were euthanized on d 21, and fibulas were analyzed for bone ash. In both experiments, pigs fed increasing inorganic P had improved (linear, $P < 0.01$) G:F and percentage bone ash. Pigs fed increasing OptiPhos had improved (Exp. 1: linear, $P < 0.001$; Exp. 2: quadratic, $P < 0.001$) percentage bone ash. Pigs fed increasing Phyzyme had improved (linear, $P < 0.001$) percentage bone ash. In Exp. 2, increasing Ronozyme P improved (quadratic, $P < 0.01$) percentage bone ash. With AOAC analyzed values and bone ash as the response variable, aP release for up to 1,000 FTU/kg of *Escherichia coli* (*E. coli*)-derived phytases (OptiPhos 2000 – M and Phyzyme XP) can be predicted by the equation ($y = -0.000000125x^2 + 0.000236245x + 0.015482$), where x is the phytase level in the diet.

Key Words: growth, nursery pig, phytase

Introduction

Phosphorus is a significant mineral in swine nutrition (Crenshaw, 2001). Cereal grains often store P in the form of phytic acid (myo-inositol hexaphosphate), called phytate (Erdman,

1979). Pigs cannot easily use P bound as phytate because they lack sufficient intestinal phytase for its liberation (Cromwell, 1980; Jongbloed et al., 1991). Adding a phytase enzyme to diets can enhance the pig's ability to hydrolyze phytate (Adeola et al., 2004). Many trials have been conducted to evaluate different sources of the phytase enzyme, including some prominent versions of the enzyme obtained from *E. coli* or *Peniophora lycii* (Adeola et al., 2006; Braña et al., 2006; Pontoppidan et al., 2007).

Comparing phytase sources and levels can be confusing because phytase manufacturers have individual analytical techniques. For instance, Augspurger et al. (2004) demonstrated that 0.13% of available P (aP) can be replaced in a corn–soybean meal-based diet with 250 phytase units (FTU)/kg OptiPhos 2000 – M (Phytex, LLC, Sheridan, IN). For Phyzyme XP 5000 G (Danisco Animal Nutrition, Marlborough, UK), 500 FTU/kg can replace 0.12% aP. Finally, 1,850 phytase units (FYT)/kg Ronozyme P – M (DSM Nutritional Products, Basel, Switzerland) is recommended to replace 0.10% aP. To avoid this confusion, the current study used inclusion rates as directed by the product labels, which gives field-applicable available P release values. To further clarify comparisons, AOAC analysis was conducted on all phytase samples (AOAC, 2000).

Additionally, phytase may be added at lower levels. However, more data is needed to determine a release curve for OptiPhos, Phyzyme XP, and Ronozyme P. Development of dose-response curves for P release could allow optimum use of different sources of the enzyme at a variety of levels.

Objectives of these trials were to evaluate the effects of three different sources of commercially-available phytase on late nursery pig performance and to develop an available P release curve.

Materials and methods

Experimental procedures used in these studies were approved by the Kansas State University Institutional Animal Care and Use Committee. In Exp. 1, a total of 88 barrows (PIC C29 × 337, Hendersonville, TN; initial BW 10.3 kg) were used in a 21-d growth trial. Pigs were blocked by weight and allotted to 1 of 11 dietary treatments. In Exp. 2, a total of 96 pigs (PIC C29 × 337, Hendersonville, TN; initial BW 9.7 kg) were used in a 21-d growth trial. Pigs were

blocked by sex and weight and allotted to 1 of 12 dietary treatments. Both experiments had 1 pig per pen and 8 pens per treatment. Each pen (0.8×1.0 m²) contained a 2-hole, dry self-feeder, and 1 nipple waterer to provide ad libitum access to feed and water. The studies were conducted in 4 adjacent rooms in the Discovery Nursery at JBS-United's Burton Russell Research Farm (Frankfurt, IN). Samples of phytase and inorganic P premixes and complete feed were taken at the time of diet preparation and analyzed for phytase concentrations.

A common starter diet (meal form) containing 0.06% aP was fed to pigs for 6 d before the experiment while pigs were being acclimated to the barn. Pigs were fed a basal diet (0.06% aP) and 2 levels of added aP monocalcium P (0.075 and 0.15 for Exp. 1 and 0.07 and 0.14 for Exp. 2) to develop a standard curve. In Exp. 1, 100, 175, 250, or 500 FTU/kg OptiPhos 2000 – M or 200, 350, 500, or 1,000 FTU/kg Phyzyme XP were added to the basal diet. In Exp. 2, 250, 500, 750, or 1,000 FTU/kg OptiPhos 2000 – M; 500, 1,000, or 1,500 FTU/kg Phyzyme XP; or 1,850 or 3,700 FYT/kg Ronozyme P - (CT) were added to the basal diet.

In Exp. 1, all treatment diets were constructed from a single basal diet (Table 1) made in two batches at the Kansas State University (**KSU**) Animal Science Feed Mill. Each bag was marked by batch and bagging order. The first 3 and last 2 bags of each batch were discarded and not used in experimental diet preparation. Individual treatments were mixed from the basal diet. A total of 89.6 kg of each batch of the basal diet were used to make 179.2 kg of each treatment diet. To mix the experimental treatments, first 44.8 kg from each of the 2 batches (a total of 89.6 kg) and was mixed for 2 min. Second, a total of 2.3 kg (0.9 kg phytase premix and 1.4 kg P premix) of premix were added to the mixer while the mixer hands were on the upside, and the diet was mixed for an additional 2 min. Third, the additional 44.8 kg of each batch of the basal diet was added (total of 89.6 kg), and the diet was mixed for an additional 2 min. Fourth, approximately 14 kg of feed were removed from the mixer discharge and deposited back into the top of the mixer. Fifth, the diet was mixed for an additional 6 min. Lastly, treatments were bagged into 13.6-kg unlined bags and tagged by treatment.

In Exp. 2, premixes were manufactured at KSU and shipped to Sheridan, IN, where they were added to a single basal diet (Table 1) that was made in 3 batches at the Burton Russell Research Farm Feed Mill (Frankfort, IN). Each bag was marked by batch and bagging order. The first and last 2 bags of each batch were discarded and not used in diet preparation. A total of

41.8, 68.9, and 68.0 kg of batch 1, 2, and 3 of the basal diet, respectively, were used to make 178.7 kg of each treatment diet. To mix the experimental treatments, first, half of each batch (total of 89.4 kg) was added to the mixer and mixed for 2 min. Second, a total of 2.7 kg (0.9 kg phytase premix and 1.8 kg inorganic P premix) of premix were added to the mixer while the mixer hands were on the upside, and the diet was mixed for an additional 2 min. Third, the remainder each batch of the basal diet was added, and the diet was mixed for an additional 2 min. Fourth, approximately 14 kg of feed were removed from the mixer discharge and deposited back into the top of the mixer. Fifth, the diet was mixed for an additional 2 min for a total treatment addition mixing time of 8 min. Lastly, treatments were bagged into 13.6-kg unlined bags and tagged by treatment.

Treatment premixes for both experiments were made at the KSU Swine Research Laboratory. The phytase premixes consisted of cornstarch with or without a phytase source (OptiPhos, Phyzyme XP, or Ronozyme P). The same lots of OptiPhos and Phyzyme XP were used to make premixes for both experiments. Phytase was stored in a freezer for approximately 6 mo. between experiments. The negative control and diets with monocalcium P were made with no phytase and 0.9 kg of cornstarch. In Exp. 1, a single batch of both the 500 FTU/kg OptiPhos premix and 1,000 FTU/kg Phyzyme XP premix was manufactured and analyzed for Lys, Ca, P, and phytase content (AOAC, 2000; Table 2). Micro ingredients were also analyzed for Ca (AOAC, 2000; Table 3). In Exp. 2, a single batch of the 1,000 FTU/kg OptiPhos premix, 1,500 FTU/kg Phyzyme XP premix, and 3,700 FYT/kg Ronozyme P premix was made and analyzed for Ca, P, and phytase content (AOAC, 2000; Table 4). Cornstarch was added in increasing levels to the base mixes to dilute them to the various phytase levels used in the trials. In both experiments, P premixes consisted of sand with or without monocalcium phosphate (21% P). The negative control and diets containing phytase were made with no monocalcium P and 1.4 (Exp. 1) or 1.8 (Exp. 2) kg of sand. Premixes were analyzed for Ca and P, and phytase analysis was conducted according to the AOAC and Phytex methods (AOAC, 2000; Table 4).

Treatment diets were fed in meal form for 21 d. Average daily gain, ADFI, and G:F were determined by weighing pigs and measuring feed disappearance on d 0 and 21 of the trial. Animals were euthanized by a licensed veterinarian via lethal injection with Euthanasia-III Solution (Exp. 1; Med-Pharmex) or Beuthanasia-D Special (Exp. 2; Schering-Plough) according

to KSU Institutional Animal Care and Use Committee standards. The right fibula was removed without cartilage caps from each animal, autoclaved, and boiled for 45 to 60 min. Fibulas were cleaned of adhering tissue, dried at 105°C for 24 h, and ashed in a muffle furnace at 600°C for 24 h. Total ash weight and percentage bone ash were measured..

Statistical Analysis

All values that were greater than 3 standard deviations away from the mean were considered outliers and removed from the data analysis. In Exp. 1, 4 pigs with outliers for growth data (ADG, ADFI, or G:F) were removed from both the growth and bone (ash weight and percentage bone ash) results. Two pigs with outliers for percentage bone ash were removed from the ash weight and percentage bone ash results but were used for calculation of growth data. One pig with an outlier for ash weight was removed from the ash weight results but was used in calculation of percentage bone ash and growth data. Three fibulas were broken during analysis, preventing ash weight and percentage bone ash for these fibulas from being calculated. Growth data from these pigs were used. In Exp. 2, 1 pig was an outlier for G:F and was removed from all data. One pig was considered an outlier for percentage bone ash and was removed from the ash weight and percentage bone ash results but used for calculation of growth data.

Data were analyzed as a randomized complete block design with pig as the experimental unit. Analysis of variance was performed with the MIXED procedure of SAS. Treatment was considered a fixed effect while pig and room were considered random effects in the model. Results were considered significant if their *P*-values were ≤ 0.05 and were considered to be a trend if their *P*-values were ≤ 0.10 . Main effects from Exp. 1 showed that the negative control and all treatments that including inorganic P remained in the linear portion of the phytase release curve. Thus, these treatments were used for generation of a standard curve predicting aP release. Standard curves were derived for ADG, G:F, ash weight, and percentage bone ash using regression analysis.

A regression equation was A standard curve was calculated for ADG, G:F, ash weight, and percentage bone ash to predict the percentage aP released from the *E. coli*-derived phytases given each response criteria. First, total intake of aP from the diet was calculated and termed to be the dosage of aP administered to each pig through its diet. Dosage for pigs fed the negative

control, OptiPhos, Phyzyme XP, and Ronozyme P diets was the product of 0.06 and individual grams of feed intake. In Exp. 1, dosage for pigs fed the negative control diet plus 0.075% aP from the monocalcium P diet was the product of 0.135 and individual grams of feed intake. Dosage for pigs fed the negative control plus 0.15% aP from the monocalcium P diet was the product of 0.21 and individual grams of feed intake. In Exp. 2, dosage for pigs fed the negative control diet plus 0.07% aP from the monocalcium P diet was the product of 0.13 and individual grams of feed intake. Dosage for pigs fed the negative control plus 0.14 aP from the monocalcium P diet was the product of 0.20 and individual grams of feed intake.

Using these aP dosages, regression was used to determine the aP release from each phytase source for a given aP dosage (intercept) and the aP release from each response variable for a given aP dosage (slope). The percentage aP released from each phytase source (y) was then calculated by adding the value of aP release from each phytase source for a given aP dosage to the product of the value of aP release from each response variable for a given aP dosage and the value of the response variable (x).

Results

Chemical analysis

In Exp. 1, Lys and P analysis of the diets resulted in concentrations similar to those used in diet formulation (Table 2). However, Ca levels were higher than expected because of higher-than-anticipated Ca levels in the micro ingredients. The high Ca levels resulted in high Ca to total P ratios (Ca:P; 2.04 to 2.20) for the negative control and all phytase diets. Lower Ca:P ratios were used in Exp. 2, in which analysis of the diets resulted in concentrations similar to those used in diet formulation.

According to AOAC analysis, the phytase concentration in OptiPhos for Exp. 1 was nearly 3.1 times the concentration listed on the label by the manufacturer, while it was 2.5 times the listed concentration in Exp. 2 (Tables 5 and 6). The phytase level in Phyzyme XP was at the concentration listed on the label by the manufacturer in Exp. 1 but 0.7 times the listed concentration in Exp. 2. Ronozyme P was only used and analyzed in Exp. 2, and analyzed values were similar to levels reported on the bag by the manufacturer.

Results of the AOAC analysis in both experiments indicated that, as expected, phytase levels increased linearly as more phytase premix was added to the diet. Phytase analysis with the Phytex assay revealed much lower phytase levels for all premixes and diets. Results from the Phytex analysis assay were not as consistent with added dietary levels as results from the AOAC assays; however, the Phytex assay was conducted by only one laboratory, whereas the AOAC assay was an average of results from three (Exp. 1) or two (Exp. 2) laboratories. Within laboratory, the Phytex assay was less consistent with calculated values than any single AOAC assay.

Experiment 1

Pigs fed increasing monocalcium P had improved (linear, $P < 0.02$) ADG, ADFI, G:F, bone ash weight, and percentage bone ash (Tables 7 and 8). Pigs fed increasing OptiPhos had improved (linear, $P < 0.03$) ADG, G:F, and percentage bone ash as well as increased (quadratic, $P = 0.05$) bone ash weight. Pigs fed increasing Phyzyme XP had improved (linear, $P < 0.04$) ADG and G:F, as well as a tendency for increased (linear, $P = 0.06$) percentage bone ash.

Percentage aP released from each phytase source varied on the basis of the response criteria used to calculate the value (Table 9). The lowest aP release value for both phytase sources was calculated with ADG as the response criteria. The aP release values calculated with G:F as the response criteria were nearly identical for all levels of OptiPhos, whereas levels generally increased with increasing Phyzyme to an overall release value that was similar for both phytase sources. The aP release values calculated from bone ash weight were similar for all levels of Phyzyme, with the exception of 500 FTU/kg. However, calculated aP release values were not as consistent for OptiPhos, as evidenced by the second-lowest phytase dose releasing the highest percentage aP. The clearest response to percentage aP release was calculated with percentage bone ash as the response criteria. As both OptiPhos and Phyzyme levels increased, calculated aP increased in a quadratic fashion to the highest phytase dose.

Experiment 2

Pigs fed increasing monocalcium P had improved (linear, $P < 0.001$) G:F and percentage bone ash, improved (quadratic, $P = 0.01$) ADFI, and a tendency for improved (linear, $P = 0.07$,

quadratic, $P = 0.09$) ADG (Tables 10 and 11). Pigs fed increasing OptiPhos had improved (linear, $P < 0.02$) ADG, G:F, and bone ash weight, increased (quadratic, $P < 0.001$) percentage bone ash, and tended to have increased (linear, $P = 0.07$) ADFI. Pigs fed increasing Phyzyme XP had improved (linear, $P < 0.001$) percentage bone ash, improved (quadratic, $P = 0.05$) G:F, and tended to have increased (linear, $P = 0.09$) bone ash weight. Pigs fed increasing Ronozyme P had improved (linear, $P < 0.01$) ADG, ADFI, and bone ash weight as well as improved (quadratic, $P < 0.04$) G:F and percentage bone ash.

Percentage aP released from each phytase source and level again varied on the basis of the response criteria used to calculate the value (Table 12). The lowest aP release value for 250 FTU/kg OptiPhos was calculated from ADG. The lowest aP release values for 500, 750, and 1,000 FTU/kg OptiPhos were calculated from bone ash weight. In contrast, the highest aP release level for all OptiPhos levels was calculated from bone ash percentage. The lowest aP release level for 500 FTU/kg Phyzyme was calculated from bone ash percentage, whereas the lowest levels for 1,000 and 1,500 FTU/kg Phyzyme were calculated from ADG. The highest aP release level for 500 FTU/kg Phyzyme was calculated from G:F, whereas the highest levels for 1,000 and 1,500 FTU/kg Phyzyme were calculated from bone ash percentage. Finally, the lowest aP release levels for 1,850 and 3,700 FTU/kg Ronozyme were calculated from bone ash weight and G:F, respectively. The highest aP release level for both Ronozyme levels was calculated from bone ash percentage.

Experiments 1 and 2

The response to various criteria can be plotted against the analyzed phytase level by using the average values of the AOAC phytase assays from both *E. coli* phytase sources.

Approximately 77% of the variation in response in percentage bone ash was explained by the analyzed phytase level in the diet (Figure 1). Similarly, a P release curve was calculated by plotting the aP released for each phytase level against the analyzed AOAC phytase level. With percentage bone ash as the response criteria, approximately 73% of the variation in aP release was explained by the analyzed phytase level in the diet (Figure 2). With AOAC analyzed values and bone ash as the response variable, aP release for up to 1,000 FTU/kg of *E. coli*-derived

phytases (OptiPhos and Phyzyme) can be predicted by the equation ($y = -0.000000125x^2 + 0.000236245x + 0.015482000$), where x is the phytase level in the diet.

Discussion

Previous research suggests that the high Ca to total P ratios in Exp. 1 likely decreased ADG and G:F (Qian et al., 1996; Hanni et al., 2005; Adeola et al., 2006). However, these ratios did not appear to affect percentage bone ash or the aP release levels calculated from percentage bone ash.

Higher phytase concentrations in the AOAC analysis compared with the Phytex analysis were expected because there are key differences between the Phytex assay used by the manufacturer of OptiPhos and the AOAC method. The Phytex assay extracts P with a 0.2M sodium citrate buffer, whereas the AOAC assay uses a 0.2M sodium acetate buffer, Tween 20, and bovine serum albumin. The Phytex assay incubation time is 15 min, and the AOAC assay incubation time is 60 min. Additionally, the color reagent used to measure the P released from phytic acid has a wavelength of 820 nm in the Phytex assay and 415 nm in the AOAC assay. Finally, the Phytex assay diafiltrates feed samples to remove high background P levels from monocalcium or dicalcium P before they are assayed, but the AOAC assay does not (AOAC, 2000; Augspurger et al., 2003).

The relative agreement between the calculated phytase levels and the AOAC assays from Exp. 1 Phyzyme XP and Exp. 2 Ronozyme is similar to findings from previous research (Adeola et al., 2004; Jendza et al., 2004, 2005). However, concentrations of OptiPhos and Phyzyme XP appeared to degrade by approximately 30% during the 180 d that they were stored at -20°C. More research is needed to determine the degradation rates of phytase stored in various environments.

Phosphorus deficiencies have been shown to negatively affect growth and percentage bone ash in pigs (Viperman et al., 1974; Mahan, 1982), which supports our findings in both experiments of linear increases in feed efficiency and percentage bone ash when monocalcium P was added. Increasing levels of phytase resulted in growth performance improvements that were similar to effects reported in other research. For instance, adding 500 FTU/kg Phyzyme XP to the control diet improved ADG by 11.3 and 12.9% in Exp. 1 and 2, respectively. Similarly,

adding 500 FTU/kg Phyzyme XP to diets deficient in aP improved ADG by 11.2 (Adeola et al., 2004), 7.5 (Braña et al., 2006), and 10.4% (Jendza et al., 2006).

The influence of *E. coli*-derived phytase source on level of percentage bone ash follows the quadratic response for aP release that has been previously reported for fungal phytase sources (Kornegay, 1996). The 77% of variation in percentage bone ash that can be explained by analyzed phytase value was the highest of any of the measured variables (63, 36, and 39% for ADG, G:F, and bone ash weight, respectively). This reinforces that percentage bone ash was the best variable to use to predict aP release and is in agreement with research that suggests bone mechanical properties more accurately indicate aP concentration levels than growth performance (Zhang et al., 2000; Augspurger et al., 2004; Jendza et al., 2006; Veum et al., 2006; James et al., 2008). In these trials, aP release values for *E. coli*-derived phytase levels predicted by using AOAC analyzed values agree largely with a previously published summary for fungal phytase sources (Kornegay, 1996), suggesting that aP release levels can be predicted from *E. coli*-derived phytases when their AOAC assayed value is less than 1,000 FTU/kg. More research needs to be conducted to further evaluate release values for higher phytase levels.

In summary, when percentage bone ash is used as the response criteria, the aP release for the phytase sources tested is similar to the manufacturers' recommendations when the products are used according to label phytase levels (0.12% for 250 FTU/kg OptiPhos, 0.10% for 500 FTU/kg Phyzyme, and 0.10% for 1,850 FTU/kg Ronozyme). When analyzed on an AOAC basis, aP release curves for the *E. coli* phytases appear to have similar release curves, at least up to 1,000 FTU/kg.

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Figures and Tables

Table 1. Diet composition (as-fed basis)¹

Item	Experiment 1	Experiment 2
Ingredient, %		
Corn	57.98	59.10
Soybean meal, 46.5% CP	34.98	35.01
Added premixes ²	0.50	0.60
Soybean oil	3.00	3.00
Limestone	1.50	0.25
Salt	0.35	0.35
Vitamin premix ³	0.25	0.25
Trace mineral premix ³	0.15	0.15
L-Lys-HCl	0.17	0.17
DL-Met	0.07	0.07
L-Thr	0.05	0.05
Medication ⁴	1.00	1.00
Total	100.00	100.00
Calculated composition		
Total Lys, %	1.34	1.34
Lys:ME ratio, g/Mcal	3.51	3.48
Standardized ileal digestibility, %		
Met:Lys	39	30
Met & Cys:Lys	58	57
Thr:Lys	64	62
Trp:Lys	20	19
Val:Lys	76	74
CP, %	21.4	21.5
ME, kcal/kg	3,450	3,459
Ca, %	0.71	0.49
P, %	0.40	0.39
Available P, %	0.06	0.06

¹ Pigs were fed experimental diets from d 0 to 21 of the trial.

² Premixes were added by hand for each treatment and consisted of 1.4 or 1.8 kg P premix.

³ Provided (per kilogram of complete diet): 11,025 IU of vitamin A; 1,654 IU of vitamin D; 44 IU of vitamin E; 4.4 mg of vitamin K (as menadione dimethylpyrimidinol bisulfate); 55.1 mg of niacin; 33.1 mg of pantothenic acid (as D-calcium pantothenate); 9.9 mg of riboflavin; 0.044 mg of vitamin B₁₂; 16.5 mg of Cu as CuSO₄·5H₂O; 165.4 mg of Fe as FeSO₄·H₂O; 39.7 mg of Mn as MnSO₄·H₂O; 0.30 mg of Se as Na₂SeO₃; 165.4 mg of Zn as ZnO; and 0.30 mg of I as C₂H₂(NH₂)₂·2HI.

⁴ Provided 5 mg/kg carbadox.

Table 2. Analyzed experimental diet nutrient composition (Exp. 1; as-fed basis)

Item	Lys, %		Ca, %		P, %		Ca:P	Phytase, FTU/kg	
	Formulated ¹	Analyzed ²	Formulated ¹	Analyzed ³	Formulated ¹	Analyzed ³	Analyzed ³	Formulated ¹	Analyzed ³
OptiPhos 2000 – M ⁴	-	0.14	-	0.10	-	0.25	-	2,000,000	5,882,333
Phyzyme XP ⁵	-	0.11	-	16.35	-	0.09	-	1,200,000	1,127,333
OptiPhos base premix ⁶	-	0.02	-	0.00	-	0.03	-	100,000	290,000
Phyzyme base premix ⁶	-	0.03	-	3.15	-	0.04	-	200,000	168,333
Negative control	1.34	1.27	0.71	0.92	0.40	0.41	2.24	-	57
0.075% aP ⁷	1.34	1.30	0.77	1.00	0.48	0.49	2.04	-	119
0.15% aP ⁷	1.34	1.25	0.84	0.90	0.55	0.58	1.55	-	77
100 FTU OptiPhos 2000 – M	1.34	1.32	0.71	0.90	0.40	0.41	2.20	100	344
175 FTU OptiPhos 2000 – M	1.34	1.34	0.71	0.98	0.40	0.41	2.39	175	560
250 FTU OptiPhos 2000 – M	1.34	1.30	0.71	0.90	0.40	0.43	2.09	250	729
500 FTU OptiPhos 2000 – M	1.34	1.37	0.71	0.95	0.40	0.43	2.21	500	1,509
200 FTU Phyzyme XP	1.34	1.32	0.71	0.93	0.40	0.43	2.16	200	213
350 FTU Phyzyme XP	1.34	1.36	0.71	1.00	0.40	0.42	2.38	350	407
500 FTU Phyzyme XP	1.34	1.31	0.71	0.92	0.40	0.43	2.14	500	429
1,000 FTU Phyzyme XP	1.34	1.30	0.71	0.97	0.40	0.43	2.26	1,000	1,038

¹ Nutrient values provided by the manufacturer.² Mean value of two samples analyzed in duplicate.³ Mean value of four samples analyzed in duplicate.⁴ Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.⁵ Danisco Animal Nutrition, Marlborough, UK.⁶ Made from the pure product and cornstarch.⁷ Added available P from monocalcium P.

Table 3. Analyzed Ca concentration of micro-ingredients (Exp. 1)

Ingredient	Analyzed, % ¹
Medication	18.18
Trace mineral premix	10.44
Vitamin premix	16.93

¹ Mean value of two samples analyzed in duplicate.

Table 4. Analyzed experimental diet nutrient composition (Exp. 2; as-fed basis)

Item	Ca, %		P, %		Ca:P	Phytase, FTU/kg	
	Formulated ¹	Analyzed ²	Formulated ¹	Analyzed ²	Analyzed ²	Formulated ¹	Analyzed ²
OptiPhos 2000 – M ³	-	0.11	-	0.02	-	2,000,000	4,946,000
Phyzyme XP ⁴	-	0.11	-	0.01	-	1,200,000	1,002,000
Ronozyme – P (M) ⁵	-	1.36	-	0.02	-	10,000,000	11,266,750
OptiPhos 2000 –M base premix ⁶	-	0.00	-	0.03	-	200,000	176,320
Phyzyme XP base premix ⁶	-	0.01	-	0.02	-	500,000	273,559
Ronozyme – P (M) premix ⁶	-	0.00	-	0.07	-	148,000	70,799
Negative control	0.49	0.48	0.39	0.36	1.33	-	58
0.07% aP ⁷	0.55	0.53	0.46	0.43	1.23	-	78
0.14% aP ⁷	0.61	0.58	0.53	0.48	1.21	-	52
250 FTU OptiPhos 2000 – M	0.49	0.53	0.39	0.36	1.47	250	674
500 FTU OptiPhos 2000 – M	0.49	0.47	0.39	0.36	1.31	500	1,227
750 FTU OptiPhos 2000 – M	0.49	0.48	0.39	0.36	1.33	750	1,849
1,000 FTU OptiPhos 2000 – M	0.49	0.49	0.39	0.36	1.36	1,000	2,479
500 FTU Phyzyme XP	0.49	0.53	0.39	0.37	1.43	500	369
1,000 FTU Phyzyme XP	0.49	0.50	0.39	0.37	1.35	1,000	708
1,500 FTU Phyzyme XP	0.49	0.47	0.39	0.37	1.27	1,500	1,091
1,850 FYT Ronozyme – P (M)	0.49	0.49	0.39	0.36	1.36	1,850	1,694
3,700 FYT Ronozyme – P (M)	0.49	0.47	0.39	0.36	1.31	3,700	3,778

¹ Nutrient values provided by the manufacturer.² Mean value of four samples analyzed in duplicate.³ Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.⁴ Danisco Animal Nutrition, Marlborough, UK.⁵ DSM Nutritional Products, Basel, Switzerland.⁶ Made from the pure product and cornstarch.⁷ Added available P from monocalcium P.

Table 5. Analyzed phytase composition (Exp. 1)

Item	Added aP from Monocalcium P			OptiPhos 2000 – M ¹ , FTU/kg				Phyzyme XP ² , FTU/kg			
	None ³	0.075%	0.15%	100	175	250	500	200	350	500	1,000
Analyzed ⁴											
AOAC assay, FTU/kg											
Laboratory A	50	70	55	335	635	740	1,635	180	465	450	1,225
Laboratory B	33	87	57	344	530	719	1,528	241	385	415	1,100
Laboratory C	88	202	119	354	516	729	1,363	219	370	423	789
Average AOAC Assay	57	119	77	344	560	729	1,509	213	407	429	1,038
Phytex assay, FTU/kg	52	86	71	275	270	300	605	225	285	280	385
Average AOAC ratio ⁵				3.5	2.9	2.9	2.7	1.1	1.1	0.8	0.8
Phytex ratio ⁶				2.8	1.5	1.2	1.2	1.1	0.8	0.6	0.4

¹ Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

² Danisco Animal Nutrition, Marlborough, UK.

³ Contained 0.06% available P.

⁴ Average of samples taken at the beginning and end of the experiment.

⁵ Ratio of AOAC analysis to formulated values.

⁶ Ratio of Phytex analysis to formulated values.

Table 6. Analyzed phytase composition (Exp. 2)

Item	Added aP from Monocalcium P			OptiPhos 2000 – M ¹ FTU/kg				Phyzyme XP ² FTU/kg			Ronozyme - P (CT) ³ FYT/kg	
	None ⁴	0.07%	0.14%	250	500	750	1,000	500	1,000	1,500	1,850	3,700
Analyzed												
AOAC assay, FTU/kg												
Laboratory A	50	50	40	710	1,330	2,000	2,600	290	760	1,140	1,790	3,920
Laboratory B	65	105	63	637	1,123	1,697	2,357	447	656	1,042	1,597	3,635
Avg. AOAC assay	58	78	52	674	1,227	1,849	2,479	369	708	1,091	1,694	3,778
Phytex assay, FTU/kg	70	84	160	360	670	800	900	180	240	550	930	1,900
Avg. AOAC ratio ⁵				2.69	2.45	2.46	2.48	0.74	0.71	0.73	0.92	1.02
Phytex ratio ⁶				1.44	1.34	1.07	0.90	0.36	0.24	0.37	0.50	0.51

¹ Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

² Danisco Animal Nutrition, Marlborough, UK.

³ DSM Nutritional Products, Basel, Switzerland.

⁴ Contained 0.06% available P.

⁵ Ratio of average AOAC analyses to formulated values.

⁶ Ratio of Phytex analyses to formulated values.

Table 7. Effects of different sources of *E-coli*-derived phytase on nursery pig performance (Exp. 1)¹

Item	Added aP from Monocalcium P			OptiPhos 2000 – M ² FTU/kg				Phyzyme XP ³ FTU/kg			
	None ⁴	0.075%	0.15%	100	175	250	500	200	350	500	1,000
d 0 to 21											
ADG, g	369	510	596	390	390	418	460	368	394	416	419
ADFI, g	744	890	888	693	718	735	791	706	762	733	729
G:F	0.51	0.57	0.67	0.56	0.56	0.57	0.58	0.52	0.52	0.57	0.58
Bone ash weight, mg	473	579	777	504	650	616	594	586	610	546	593
Bone ash, %	35.6	39.4	41.8	36.2	38.2	39.6	41.1	37.0	39.0	37.9	40.0

¹ A total of 88 pigs (1 pig per pen and 8 pens per treatment) with an initial BW of 10.3 kg. Pigs were fed the control diet (0.06% aP) during a 6-d pretest period then fed experimental diets for 21 d.

² Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

³ Danisco Animal Nutrition, Marlborough, UK.

⁴ Contained 0.06% available P.

Table 8. Main effects of different sources of *E-coli*-derived phytase on nursery pig performance (Exp. 1)¹

Item	Monocalcium P		OptiPhos 2000 – M ²		Phyzyme XP ³		SE
	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	
d 0 to 21							
ADG, g	0.0001	0.25	0.001	0.90	0.03	0.49	20.7
ADFI, g	0.004	0.07	0.11	0.22	0.91	0.90	34.2
G:F	0.0001	0.54	0.02	0.14	0.01	0.64	0.023
Bone ash weight, mg	0.01	0.47	0.07	0.05	0.27	0.30	55.0
Bone ash, %	0.01	0.69	0.01	0.56	0.06	0.59	1.65

¹ A total of 88 pigs (1 pig per pen and 8 pens per treatment) with an initial BW of 10.3 kg. Pigs were fed the control diet (0.06% aP) during a 6-d pretest period then fed experimental diets for 21 d.

² Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

³ Danisco Animal Nutrition, Marlborough, UK.

Table 9. Calculated available P release values based on different response criteria (Exp. 1)

Item	OptiPhos 2000 – M ¹ , FTU/kg				Phyzyme XP ² , FTU/kg				SE
	100	175	250	500	200	350	500	1,000	
ADG	0.029	0.029	0.046	0.063	0.013	0.022	0.042	0.044	0.012
G:F	0.099	0.096	0.097	0.089	0.068	0.056	0.093	0.102	0.018
Bone ash weight	0.055	0.127	0.105	0.084	0.092	0.094	0.070	0.094	0.028
Bone ash	0.059	0.086	0.117	0.121	0.069	0.094	0.082	0.120	0.028

¹ Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

² Danisco Animal Nutrition, Marlborough, UK.

Table 10. Effects of different sources of *E-coli*-derived phytase on nursery pig performance (Exp. 2)¹

Item	Additional aP from Monocalcium P			OptiPhos 2000 – M ² , FTU/kg				Phyzyme XP ³ , FTU/kg			Ronozyme - P (CT) ⁴ , FTU/kg	
	None ⁵	0.07%	0.14%	250	500	750	1,000	500	1,000	1,500	1,850	3,700
d 0 to 21												
ADG, g	404	487	469	477	501	520	520	464	452	444	482	578
ADFI, g	647	769	677	718	734	749	737	678	672	647	692	831
G:F	0.63	0.64	0.69	0.66	0.68	0.69	0.71	0.69	0.67	0.68	0.70	0.70
Bone ash weight, mg	626	601	696	731	734	744	799	625	773	681	691	799
Bone ash, %	34.2	39.6	41.2	41.6	41.9	42.7	43.6	37.1	41.9	42.0	41.1	42.3

¹ A total of 128 pigs (1 pig per pen and 8 pens per treatment) with an initial BW of 9.7 kg. Pigs were fed the control diet (0.06% aP) during a 6-d pretest period, then fed experimental diets for 21 d.

² Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN

³ Danisco Animal Nutrition, Marlborough, UK.

⁴ DSM Nutritional Products, Basel, Switzerland.

⁵ Contained 0.06% available P.

Table 11. Main effects of different sources of *E-coli*-derived phytase on nursery pig performance (Exp. 2)¹

Probabilities, <i>P</i> <									
Item	Added Monocalcium P		OptiPhos 2000 – M ²		Phyzyme XP ³		Ronozyme – P (CT) ⁴		SE
	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	
d 0 to 21									
ADG, g	0.07	0.09	0.001	0.11	0.33	0.18	0.001	0.76	29.7
ADFI, g	0.54	0.01	0.07	0.21	0.96	0.43	0.001	0.28	43.2
G:F	0.001	0.19	0.001	0.24	0.01	0.05	0.001	0.03	0.014
Bone ash weight, mg	0.23	0.26	0.01	0.56	0.09	0.28	0.004	0.67	60.2
Bone ash, %	0.001	0.07	0.001	0.001	0.001	0.10	0.001	0.01	1.21

¹ A total of 128 pigs (1 pig per pen and 8 pens per treatment) with an initial BW of 9.7 kg. Pigs were fed the control diet (0.06% aP) during a 6-d pretest period, then fed experimental diets for 21 d.

² Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

³ Danisco Animal Nutrition, Marlborough, UK.

⁴ DSM Nutritional Products, Basel, Switzerland.

Table 12. Effects of different sources of *E-coli*-derived phytase on nursery pig aP release (Exp. 2)¹

Item	OptiPhos 2000 – M ² , FTU/kg				Phyzyme XP ³ , FTU/kg			Ronozyme - P (CT) ⁴ , FTU/kg		SE
	250	500	750	1,000	500	1,000	1,500	1,850	3,700	
Predicted aP, %										
ADG	0.075	0.084	0.090	0.093	0.079	0.072	0.073	0.084	0.098	0.008
G:F	0.079	0.082	0.082	0.092	0.098	0.095	0.099	0.097	0.070	0.015
Bone ash weight	0.088	0.079	0.079	0.091	0.072	0.104	0.090	0.081	0.074	0.012
Bone ash, %	0.127	0.115	0.125	0.142	0.056	0.137	0.146	0.117	0.103	0.021

¹ A total of 128 pigs (1 pig per pen and 8 pens per treatment) with an initial BW of 9.7 kg. Pigs were fed the control diet (0.06% aP) during a 6-d pretest period, then fed experimental diets for 21 d.

² Phytex LLC, a majority owned subsidiary of JBS United, Inc., Sheridan, IN.

³ Danisco Animal Nutrition, Marlborough, UK.

⁴ DSM Nutritional Products, Basel, Switzerland.

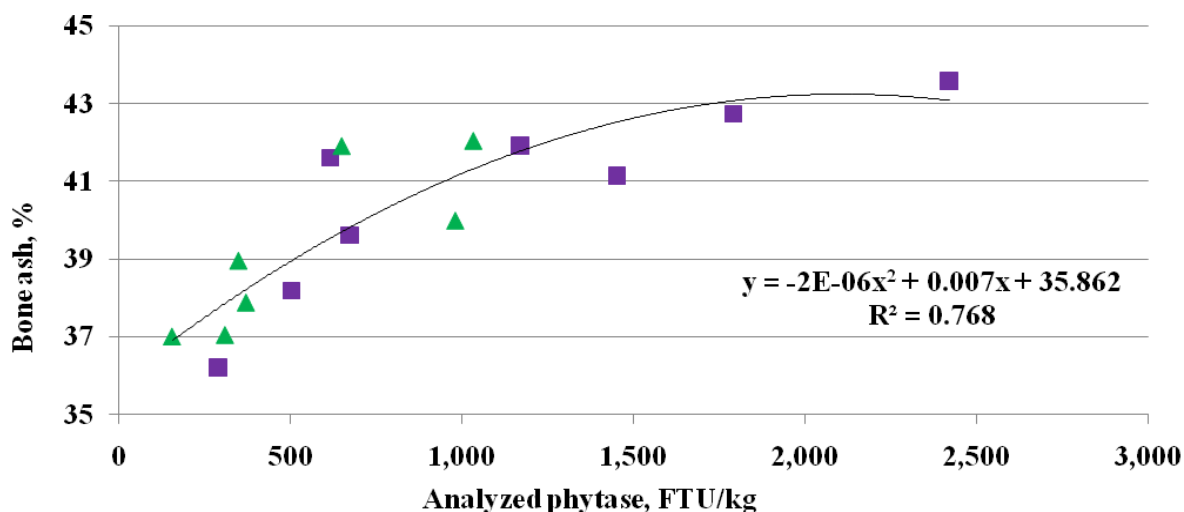


Figure 1. Analyzed phytase level vs. percentage bone ash. Percentage bone ash and average values from AOAC phytase assays of OptiPhos 2000 – M (■) and Phyzyme XP (▲) explain that 76.8% of the variation in percentage bone ash can be determined by analyzed *E. coli*-derived phytase levels in the diet.

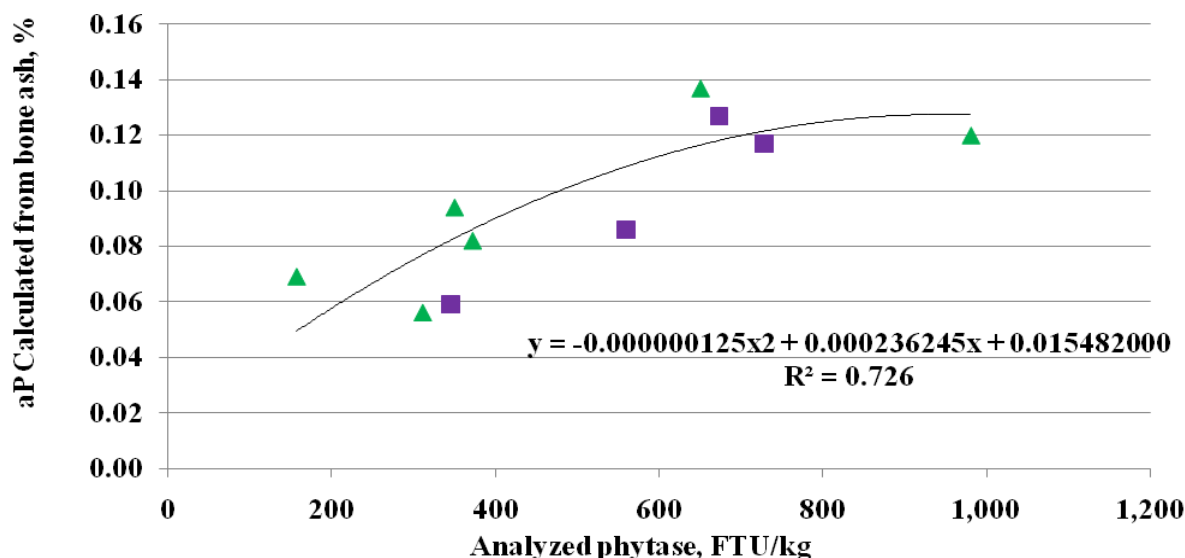


Figure 2. Analyzed phytase level vs. aP calculated from percentage bone ash. The calculated level of available P (aP) released from percentage bone ash and the average values from AOAC phytase assays of OptiPhos 2000 – M (■) and Phyzyme XP (▲) explain that 72.6% of the variation in the calculated level of aP released from percentage bone ash can be determined by analyzed *E. coli*-derived phytase levels in the diet. Additionally, aP release can be predicted by the equation ($y = -0.000000125x^2 + 0.000236245x + 0.015482000$), where x is the phytase level in the diet.